Sox2 and Cancer Biology Research Literatures

Mark Herbert

Queens, NY 11418, USA, ma8080@gmail.com

Abstract: Cancer is the general name for a group of more than 100 diseases. Although there are many kinds of cancer, all cancers start because abnormal cells grow out of control. Untreated cancers can cause serious illness and death. The body is made up of trillions of living cells. Normal body cells grow, divide, and die in an orderly fashion. During the early years of a person's life, normal cells divide faster to allow the person to grow. After the person becomes an adult, most cells divide only to replace worn-out or dying cells or to repair injuries. This article introduces recent research reports as references in the related studies.
[Herbert M. **Sox2 and Cancer Biology Research Literatures.** *Academ Arena* 2019;11(2):16-71]. ISSN 1553-992X (print); ISSN 2158-771X (online). http://www.sciencepub.net/academia. 4. doi:10.7537/marsaaj110219.04.

Key words: cancer; life; research; literature; cell

1. Introduction

Cancer is the general name for a group of more than 100 diseases. Although there are many kinds of cancer, all cancers start because abnormal cells grow out of control. Untreated cancers can cause serious illness and death. The body is made up of trillions of living cells. Normal body cells grow, divide, and die in an orderly fashion. During the early years of a person's life, normal cells divide faster to allow the person to grow. After the person becomes an adult, most cells divide only to replace worn-out or dying cells or to repair injuries.

The following introduces recent reports as references in the related studies.

Abd El-Maqsoud, N. M. and D. M. Abd El-Rehim (2014)."Clinicopathologic implications of EpCAM and Sox2 expression in breast cancer."Clin Breast Cancer**14**(1): e1-9.

BACKGROUND: The purpose of this study was to investigate the clinicopathologic significance of EpCAM and Sox2 expression in breast cancer and to study their correlation during breast cancer progression. PATIENTS AND METHODS: EpCAm and Sox2 expression were assessed using immunohistochemistry in ductal carcinoma insitu (DCIS), invasive breast cancer (IBC) and matched lymph node metastasis (LNM), if present. RESULTS: EpCAM overexpression was found in 63.2% of DCIS, 72.2% of IBC and 74.4% of LNM. In IBC cases, EpCAM overexpression was associated with high grade (P <.001), large tumor size (P =.051), poor Nottingham Prognostic Index (NPI) (P =.006), histological tumor types (P =.044) and the triple negative phenotype (P =.008). LNM frequently reflected the expression phenotype of the matched primary tumors with no significant differences between LNM and their primary tumors (P =.564). Sox2 expression was detected in 47.4%, 33.3% and 54.7% of DCIS, IBC and LNM respectively. In DCIS group, Sox2 expression was significantly associated with comedo type (P =.037), negative ER (P =.012) and PR (P =.037) and the triple negative phenotype (P =.006). In IBC cases, Sox2 expression showed significant associations with high grade (P =.045), nodal spread (P =.037), poor NPI (P =.018) and the triple negative phenotype (P <.001). LNM showed significantly higher Sox2 expression rates than primary tumors (P <.001). Significant positive associations between EpCAM overexpression and Sox2 positivity in DCIS (P =.027), IBC (P =.001) and LNM (P <.001) were found. CONCLUSION: This study emphasized the potential role of EpCAM and Sox2 in breast carcinogenesis and revealed their involvement during breast cancer progression and LN metastases.

Akimoto, N., et al. (2018). "Transfection of T-Box Transcription Factor BRACHYURY and SOX2 Synergistically Promote Self-Renewal and Invasive Phenotype in Oral Cancer Cells." Int J Mol Sci**19**(11).

Recent studies suggest that epithelial (-)mesenchymal transition (EMT) correlates with cancer metastasis. In addition, there is growing evidence of the association of EMT with cancer stem cells (CSCs). Recently, we showed that the T-box transcription factor BRACHYURY could be a strong regulator of EMT and the CSC phenotype, which were effectively suppressed by a BRACHYURY knockdown in an adenoid cystic carcinoma cell line. In this study, we further tested whether BRACHYURY is a regulator of cancer stemness by means of forced expression of BRACHYURY in oral cancer cell lines. BRACHYURY, SOX2, or both were stably transfected into oral carcinoma cell lines. We analysed these transfectants with respect to self-renewal phenotypes using a sphere-formation assay, and we assessed the expression levels of EMT markers and

stem cell markers using real-time reverse transcription-polymerase chain reaction (RT-PCR). Cell migration and invasiveness in vitro were evaluated using a wound healing assay and a tumour cell dissemination assay, respectively. Forced expression of BRACHYURY or SOX2 slightly increased expression of EMT and stem cell markers and the self-renewal phenotype. The expression levels, however, were much lower compared to those of cancer stem cell-like cells. Forced co-expression of BRACHYURY and SOX2 strongly upregulated EMT and stem cell markers and the self-renewal phenotype. Cell migration and invasiveness in vitro were also remarkably enhanced. These synergistic effects increased expression levels of FIBRONECTIN, SNAIL, SLUG, ZEB1, and TGF-beta2. In particular, the effects on FIBRONECTIN and TGF-beta2 were significant. We found that BRACHYURY and SOX2 synergistically promote cancer stemness in oral cancer cells. This finding points to the importance of gene or protein networks associated with BRACHYURY and SOX2 in the development and maintenance of the CSC phenotype.

Alqahtani, H., et al. (2016). "DDX17 (P72), a Sox2 binding partner, promotes stem-like features conferred by Sox2 in a small cell population in estrogen receptor-positive breast cancer." Cell Signal**28**(2): 42-50.

We have previously demonstrated the existence of two phenotypically distinct cell subsets in estrogen receptor (ER)-positive breast cancer (BC) based on their differential response to a Sox2 reporter (SRR2), with reporter responsive (RR) cells being more tumorigenic and stem-like than reporter unresponsive (RU) cells. To delineate the molecular mechanisms underlying this phenotypic dichotomy, we tested our hypothesis that Sox2, which is a key regulator of the RR phenotype, is under the control of its binding partners. In this study, we focused on DDX17, known to be a transcription co-activator and found to be a Sox2 binding partner by liquid chromatography-mass spectrometry. Using immunoprecipitation, we confirmed the binding between DDX17 and Sox2, although this interaction was largely restricted to RR cells. While DDX17 was found in both the cytoplasm and nuclei in RU cells, it is confined to the nuclei in RR cells. siRNA knockdown of DDX17 in RR cells substantially decreased the Sox2-SRR2 binding and significantly decreased the SRR2 reporter activity without affecting the protein level of Sox2. Using ChIP-PCR, DDX17 knockdown also significantly decreased the binding of Sox2 to genomic SRR2, as well as 3 of its specific gene targets including MUC15, CCND1 and CD133. Correlating with these findings, siRNA knockdown of DDX17 significantly reduced soft agar colony formation and mammosphere formation in RR cells but not RU cells. To conclude, DDX17 is a Sox2-binding protein in ER-positive BC. In RR but not RU cells, DDX17 enhances the tumorigenic and stem-like features of Sox2 by promoting its binding to its target genes.

Amini, S., et al. (2014). "The expressions of stem cell markers: Oct4, Nanog, Sox2, nucleostemin, Bmi, Zfx, Tcl1, Tbx3, Dppa4, and Esrrb in bladder, colon, and prostate cancer, and certain cancer cell lines." Anat Cell Biol**47**(1): 1-11.

Uncontrolled self-renewal plays a direct function in the progression of different types of carcinomas. The same molecular pathway that manages self-renewal in normal stem cells also seems to manage cancer stem cells. Here, we examine the expressions of self-renewal regulatory factors Oct4, Nanog, Sox2, nucleostemin, Zfx, Esrrb, Tcl1, Tbx3, and Dppa4 in tissue samples of colon, prostate, and bladder carcinomas as well as cancer cell lines HT-29, Caco-2, HT-1376, LNCaP, and HepG2. We used reverse transcriptase polymerase chain reaction to examine expressions of the above mentioned regulatory factors in cancer cell lines HT-29, Caco-2, HT-1376, LNCaP, and HepG2 and in 20 tumor tissue samples. Total RNA was isolated by the ISOGEN method. RNA integrity was checked by agarose gel electrophoresis and spectrophotometry. Expressions of Oct4 and nucleostemin at the protein level were determined by immunocytochemistry. A significant relationship was found between tumor grade and self-renewal gene expression. Expressions of stem cell specific marker genes were detected in all examined cancer cell lines, in 40% to 100% of bladder cancer samples, and in 60% to 100% of colon and prostate cancer samples. Oct4 expressed in 100% of tumor tissue samples. Our data show that stem cell markers Oct4, Nanog, Sox2, nucleostemin, Bmi, Zfx, Esrrb, Tcl1, Tbx3, and Dppa4 significantly express in cancer cell lines and cancer tissues. Hence, these markers might be useful as potential tumor markers in the diagnosis and/or prognosis of tumors.

Askarian-Amiri, M. E., et al. (2014). "Emerging role of long non-coding RNA SOX2OT in SOX2 regulation in breast cancer."PLoS One**9**(7): e102140.

The transcription factor SOX2 is essential for maintaining pluripotency in a variety of stem cells. It has important functions during embryonic development, is involved in cancer stem cell maintenance, and is often deregulated in cancer. The mechanism of SOX2 regulation has yet to be clarified, but the SOX2 gene lies in an intron of a long multi-exon non-coding RNA called SOX2 overlapping transcript (SOX2OT). Here, we show that the

expression of SOX2 and SOX2OT is concordant in breast cancer, differentially expressed in estrogen receptor positive and negative breast cancer samples and that both are up-regulated in suspension culture conditions that favor growth of stem cell phenotypes. Importantly, ectopic expression of SOX2OT led to an almost 20-fold increase in SOX2 expression, together with a reduced proliferation and increased breast cancer cell anchorage-independent growth. We propose that SOX2OT plays a key role in the induction and/or maintenance of SOX2 expression in breast cancer.

Atakan, S., et al. (2014). "Autologous anti-SOX2 antibody responses reflect intensity but not frequency of antigen expression in small cell lung cancer." BMC Clin Pathol**14**: 24.

BACKGROUND: Anti-SOX2 antibody responses are observed in about 10 to 20% of small cell lung cancer (SCLC) patients. The aim of this study was to determine whether such responses reflect a particular pattern of SOX2 protein expression in the tumor and whether this pattern associates with clinical outcome. METHODS: Paraffin embedded tumor tissues, obtained from SCLC patients who had no evidence of paraneoplastic autoimmune degeneration, were evaluated for SOX2 expression by immunohistochemistry for both intensity and extent of staining. Sera from the same patients were tested for autologous antibodies against recombinant SOX2 by enzyme-linked immunosorbent assay (ELISA). Correlates between overall survival and various clinical parameters including SOX2 staining and serology were determined. RESULTS: SOX2 protein expression was observed in tumor tissue in 89% of patients. Seventeen patients (29%) were seropositive for SOX2 antibodies and, in contrast to SOX2 staining, the presence of antibody correlated with limited disease stage ($p = 0.05$). SOX2 seropositivity showed a significant association with the intensity of SOX2 staining in the tumor ($p = 0.02$) but not with the frequency of SOX2 expressing cells. CONCLUSION: Anti-SOX2 antibodies associate with better prognosis (limited stage disease) while SOX2 protein expression does not; similar to reports from some earlier studies. Our data provides an explanation for this seemingly contrasting data for the first time as SOX2 antibodies can be observed in patients whose tumors contain relatively few but strongly staining cells, thus supporting the possible presence of active immune-surveillance and immune-editing targeting SOX2 protein in this tumor type.

Bae, K. M., et al. (2016). "Hypoxia regulates SOX2 expression to promote prostate cancer cell invasion and sphere formation." Am J Cancer Res**6**(5): 1078-1088.

SOX2 is an embryonic stem cell marker that in prostate cancer has been associated not only with tumorigenesis but also metastasis. Furthermore hypoxia in primary tumors has been linked to poor prognosis and outcomes in this disease. The goal of the present study was to investigate the impact of hypoxia on SOX2 expression and metastasis-associated functions in prostate cancer cells. A tissue microarray of 80 samples from prostate cancer patients or healthy controls was employed to examine the expression of HIF-1alpha and its correlation with SOX2. The role of SOX2 and HIF-1/2alpha in the regulation of cell invasion and sphere formation capacity under hypoxic conditions was investigated in vitro using short hairpin RNA (shRNA)-mediated knockdown in three human prostate cancer cell lines. HIF-1alpha expression was significantly elevated in malignant prostate tissue compared to benign or normal tissue, and in tumor samples its expression was highly correlated with SOX2. In prostate cancer cells, acute and chronic exposures to hypoxia that resulted in elevated expression levels of HIF-1alpha and HIF-2alpha, respectively, also induced SOX2. Genetic depletion of SOX2 attenuated hypoxia-induced cell functions. Knockdown of HIF-1alpha, but not HIF-2alpha, decreased acute hypoxia-mediated cell invasion and SOX2 up-regulation, whereas only HIF-2alpha gene silencing reduced sphere formation capacity and chronic hypoxia-mediated SOX2 up-regulation. Enhanced SOX2 expression and HIF-1alpha or HIF-2alpha associated phenotypes are dependent on the time duration of exposure to hypoxia. The present results indicate that SOX2 may be a key mediator of hypoxia-induced metastasis-associated functions and hence may serve as a potential target for therapeutic interventions for metastatic prostate cancer.

Basu-Roy, U., et al. (2015). "Sox2 antagonizes the Hippo pathway to maintain stemness in cancer cells." Nat Commun**6**: 6411.

The repressive Hippo pathway has a profound tumour suppressive role in cancer by restraining the growth-promoting function of the transcriptional coactivator, YAP. We previously showed that the stem cell transcription factor Sox2 maintains cancer stem cells (CSCs) in osteosarcomas. We now report that in these tumours, Sox2 antagonizes the Hippo pathway by direct repression of two Hippo activators, Nf2 (Merlin) and WWC1 (Kibra), leading to exaggerated YAP function. Repression of Nf2, WWC1 and high YAP expression marks the CSC fraction of the tumor population, while the more differentiated fraction has high Nf2, high WWC1 and reduced YAP expression.

YAP depletion sharply reduces CSCs and tumorigenicity of osteosarcomas. Thus, Sox2 interferes with the tumour-suppressive Hippo pathway to maintain CSCs in osteosarcomas. This Sox2-Hippo axis is conserved in other Sox2-dependent cancers such as glioblastomas. Disruption of YAP transcriptional activity could be a therapeutic strategy for Sox2-dependent tumours.

Belotte, J., et al. (2015). "Sox2 gene amplification significantly impacts overall survival in serous epithelial ovarian cancer." Reprod Sci**22**(1): 38-46.

Epithelial ovarian cancer (EOC) is the deadliest gynecologic cancer. Recently, the existence of ovarian cancer stem cells has been reported. Sox2, Nanog and Oct4 are key markers of "stemness". The objective of this study was to determine whether Sox2, Nanog, and Oct4 are associated with EOC and poor outcome. The expression of these markers was assessed by immunofluorescence staining and real-time RT-PCR in human EOC cell lines MDAH-2774 and SKOV-3, while the cancer genome atlas (TCGA) dataset was analyzed for associations with survival. Sox2, Nanog and Oct4 (POU5F1) were all detected by immunofluorescence staining and these results were confirmed by real-time RT-PCR. The TCGA dataset revealed a 26%, 9%, and 6% amplification of Sox2, Nanog and POU5F1, respectively. Additionally, K-M survival analyses showed a significant median overall survival difference (41 versus 48.3 months, P =.01) for Sox2 amplification, but not for Nanog (44.1 versus 36.2 months, P >.05) and POU5F1 (43.5 versus 45.0 months, P >.05). Our results suggest that Sox2 gene amplification significantly influences overall survival.

Bora-Singhal, N., et al. (2015). "Gli1-Mediated Regulation of Sox2 Facilitates Self-Renewal of Stem-Like Cells and Confers Resistance to EGFR Inhibitors in Non-Small Cell Lung Cancer." Neoplasia**17**(7): 538-551.

Non-small cell lung cancer (NSCLC) patients have very low survival rates because the current therapeutic strategies are not fully effective. Although EGFR tyrosine kinase inhibitors are effective for NSCLC patients harboring EGFR mutations, patients invariably develop resistance to these agents. Alterations in multiple signaling cascades have been associated with the development of resistance to EGFR inhibitors. Sonic Hedgehog and associated Gli transcription factors play a major role in embryonic development and have recently been found to be reactivated in NSCLC, and elevated Gli1 levels correlate with poor prognosis. The Hedgehog pathway has been implicated in the functions of cancer stem cells, although the underlying molecular mechanisms are not clear. In this context, we demonstrate that Gli1 is a strong regulator of embryonic stem cell transcription factor Sox2. Depletion of Gli1 or inhibition of the Hedgehog signaling significantly abrogated the self-renewal of stem-like side-population cells from NSCLCs as well as vascular mimicry of such cells. Gli1 was found to transcriptionally regulate Sox2 through its promoter region, and Gli1 could be detected on the Sox2 promoter. Inhibition of Hedgehog signaling appeared to work cooperatively with EGFR inhibitors in markedly reducing the viability of NSCLC cells as well as the self-renewal of stem-like cells. Thus, our study demonstrates a cooperative functioning of the EGFR signaling and Hedgehog pathways in governing the stem-like functions of NSCLC cancer stem cells and presents a novel therapeutic strategy to combat NSCLC harboring EGFR mutations.

Bornschein, J., et al. (2013). "Dysregulation of CDX1, CDX2 and SOX2 in patients with gastric cancer also affects the non-malignant mucosa." J Clin Pathol**66**(9): 819-822.

The interplay between gastric and intestinal transcription factors has an important impact on gastric carcinogenesis. We compared the gene expression of CDX1, CDX2, SOX2 and related downstream genes in tumour and tumour surrounding gastric tissue of 48 gastric cancer patients with 30 healthy controls. There was no difference of gene expression of CDX1 and CDX2 between tumour or tumour-adjacent and tumour-distant mucosa, but both factors were significantly higher expressed in cancer patients compared with controls ($p<0.01$). SOX2 was downregulated in tumour tissue compared to controls, whereas tumour-adjacent and tumour-distant mucosa showed intermediate SOX2 expression. Lauren type and Helicobacter pylori infection had no significant impact on expression of the transcription factors. Expression of CDX1 and CDX2 was higher in the presence of intestinal metaplasia. The differential regulation of the gene expression of CDX1, CDX2 and SOX2 in patients with gastric cancer affects not only the tumour but also the non-neoplastic tumour-distant mucosa.

Boumahdi, S., et al. (2014). "SOX2 controls tumour initiation and cancer stem-cell functions in squamous-cell carcinoma." Nature**511**(7508): 246-250.

Cancer stem cells (CSCs) have been reported in various cancers, including in skin squamous-cell carcinoma (SCC). The molecular mechanisms regulating tumour initiation and stemness are still poorly characterized. Here we find that Sox2, a transcription factor expressed in various types of

embryonic and adult stem cells, was the most upregulated transcription factor in the CSCs of squamous skin tumours in mice. SOX2 is absent in normal epidermis but begins to be expressed in the vast majority of mouse and human pre-neoplastic skin tumours, and continues to be expressed in a heterogeneous manner in invasive mouse and human SCCs. In contrast to other SCCs, in which SOX2 is frequently genetically amplified, the expression of SOX2 in mouse and human skin SCCs is transcriptionally regulated. Conditional deletion of Sox2 in the mouse epidermis markedly decreases skin tumour formation after chemical-induced carcinogenesis. Using green fluorescent protein (GFP) as a reporter of Sox2 transcriptional expression (SOX2-GFP knock-in mice), we showed that SOX2-expressing cells in invasive SCC are greatly enriched in tumour-propagating cells, which further increase upon serial transplantations. Lineage ablation of SOX2-expressing cells within primary benign and malignant SCCs leads to tumour regression, consistent with the critical role of SOX2-expressing cells in tumour maintenance. Conditional Sox2 deletion in pre-existing skin papilloma and SCC leads to tumour regression and decreases the ability of cancer cells to be propagated upon transplantation into immunodeficient mice, supporting the essential role of SOX2 in regulating CSC functions. Transcriptional profiling of SOX2-GFP-expressing CSCs and of tumour epithelial cells upon Sox2 deletion uncovered a gene network regulated by SOX2 in primary tumour cells in vivo. Chromatin immunoprecipitation identified several direct SOX2 target genes controlling tumour stemness, survival, proliferation, adhesion, invasion and paraneoplastic syndrome. We demonstrate that SOX2, by marking and regulating the functions of skin tumour-initiating cells and CSCs, establishes a continuum between tumour initiation and progression in primary skin tumours.

Bourguignon, L. Y., et al. (2012). "Hyaluronan-CD44v3 interaction with Oct4-Sox2-Nanog promotes miR-302 expression leading to self-renewal, clonal formation, and cisplatin resistance in cancer stem cells from head and neck squamous cell carcinoma." J Biol Chem**287**(39): 32800-32824.

Human head and neck squamous cell carcinoma (HNSCC) is a highly malignant cancer associated with major morbidity and mortality. In this study, we determined that human HNSCC-derived HSC-3 cells contain a subpopulation of cancer stem cells (CSCs) characterized by high levels of CD44v3 and aldehyde dehydrogenase-1 (ALDH1) expression. These tumor cells also express several stem cell markers (the transcription factors Oct4, Sox2, and Nanog) and display the hallmark CSC properties of self-renewal/clonal formation and the ability to generate heterogeneous cell populations. Importantly, hyaluronan (HA) stimulates the CD44v3 (an HA receptor) interaction with Oct4-Sox2-Nanog leading to both a complex formation and the nuclear translocation of three CSC transcription factors. Further analysis reveals that microRNA-302 (miR-302) is controlled by an upstream promoter containing Oct4-Sox2-Nanog-binding sites, whereas chromatin immunoprecipitation (ChIP) assays demonstrate that stimulation of miR-302 expression by HA-CD44 is Oct4-Sox2-Nanog-dependent in HNSCC-specific CSCs. This process results in suppression of several epigenetic regulators (AOF1/AOF2 and DNMT1) and the up-regulation of several survival proteins (cIAP-1, cIAP-2, and XIAP) leading to self-renewal, clonal formation, and cisplatin resistance. These CSCs were transfected with a specific anti-miR-302 inhibitor to silence miR-302 expression and block its target functions. Our results demonstrate that the anti-miR-302 inhibitor not only enhances the expression of AOF1/AOF2 and DNMT1 but also abrogates the production of cIAP-1, cIAP-2, and XIAP and HA-CD44v3-mediated cancer stem cell functions. Taken together, these findings strongly support the contention that the HA-induced CD44v3 interaction with Oct4-Sox2-Nanog signaling plays a pivotal role in miR-302 production leading to AOF1/AOF2/DNMT1 down-regulation and survival of protein activation. All of these events are critically important for the acquisition of cancer stem cell properties, including self-renewal, clonal formation, and chemotherapy resistance in HA-CD44v3-activated head and neck cancer.

Cai, C., et al. (2013). "[Expression of SOX2 in cervical intraepithelial neoplasia and cervical cancer and its clinical significance]."Nan Fang Yi Ke Da Xue Xue Bao**33**(1): 128-130.

OBJECTIVE: To investigate the expression of SOX2 in cervical intraepithelial neoplasia (CIN) and cervical cancer and explore its association with the clinical features. METHODS: SOX2 expressions were examined using immunohistochemical method in 10 normal cervical tissue specimens, 36 cervical intraepithelial neoplasia specimens (including 10 cases of grade I, 12 of grade II, and 14 grade III) and 40 cervical cancer specimens (including 21 cases of stage I and 19 of stage II). The correlation between the immunohistochemical results and the clinical features of the patients was analyzed. RESULTS: SOX2 expression was negative in normal cervical tissues, and was positive in 41.6% of CIN specimens (10.0% in CIN I, 41.7% in CIN II, and 64.3% in CIN III) in 82.5% of cervical cancer specimens (78.2% in stage I and 88.2% in stage II). The patients with cervical cancer had a significantly higher positivity rate of

SOX2 than normal control group (P<0.05). The positivity rate of SOX2 increased with the evolution of cervical disease. SOX2 protein expression was significantly correlated with the histological grade and lymph node metastasis (P<0.05), but not with the age or clinical stage of the patients (P<0.05). CONCLUSION: SOX2 expression may serve as a useful indicator for evaluating metastasis and malignancy of cervical cancer.

Cai, C. F., et al. (2015). "[Expression of SOX2 in cervical intraepithelial neoplasia and cervical cancer and its clinical significance]."Nan Fang Yi Ke Da Xue Xue Bao**35**(1): 128-130.

OBJECTIVE: To investigate the expression of SOX2 in cervical intraepithelial neoplasia (CIN) and cervical cancer and explore its association with the clinical features. METHODS: SOX2 expressions were examined using immunohistochemical method in 10 normal cervical tissue specimens, 36 cervical intraepithelial neoplasia specimens (including 10 cases of grade I, 12 of grade II, and 14 grade III) and 40 cervical cancer specimens (including 21 cases of stage I and 19 of stage II). The correlation between the immunohistochemical results and the clinical features of the patients was analyzed. RESULTS: SOX2 expression was negative in normal cervical tissues, and was positive in 41.6% of CIN specimens (10.0% in CIN I, 41.7% in CIN II, and 64.3% in CIN III) in 82.5% of cervical cancer specimens (78.2% in stage I and 88.2% in stage II). The patients with cervical cancer had a significantly higher positivity rate of SOX2 than normal control group (P<0.05). The positivity rate of SOX2 increased with the evolution of cervical disease. SOX2 protein expression was significantly correlated with the histological grade and lymph node metastasis (P<0.05), but not with the age or clinical stage of the patients (P<0.05). CONCLUSION: SOX2 expression may serve as a useful indicator for evaluating metastasis and malignancy of cervical cancer.

Camilo, V., et al. (2014). "Immunohistochemical molecular phenotypes of gastric cancer based on SOX2 and CDX2 predict patient outcome." BMC Cancer**14**: 753.

BACKGROUND: Gastric cancer remains a serious health concern worldwide. Patients would greatly benefit from the discovery of new biomarkers that predict outcome more accurately and allow better treatment and follow-up decisions. Here, we used a retrospective, observational study to assess the expression and prognostic value of the transcription factors SOX2 and CDX2 in gastric cancer. METHODS: SOX2, CDX2, MUC5AC and MUC2 expression were assessed in 201 gastric tumors by immunohistochemistry. SOX2 and CDX2 expression were crossed with clinicopathological and follow-up data to determine their impact on tumor behavior and outcome. Moreover, SOX2 locus copy number status was assessed by FISH (N = 21) and Copy Number Variation Assay (N = 62). RESULTS: SOX2 was expressed in 52% of the gastric tumors and was significantly associated with male gender, T stage and N stage. Moreover, SOX2 expression predicted poorer patient survival, and the combination with CDX2 defined two molecular phenotypes, SOX2+CDX2- versus SOX2-CDX2+, that predict the worst and the best long-term patients' outcome. These profiles combined with clinicopathological parameters stratify the prognosis of patients with intestinal and expanding tumors and in those without signs of venous invasion. Finally, SOX2 locus copy number gains were found in 93% of the samples reaching the amplification threshold in 14% and significantly associating with protein expression. CONCLUSIONS: We showed, for the first time, that SOX2 combined with CDX2 expression profile in gastric cancer segregate patients into different prognostic groups, complementing the clinicopathological information. We further demonstrate a molecular mechanism for SOX2 expression in a subset of gastric cancer cases.

Carina, V., et al. (2013). "Multiple pluripotent stem cell markers in human anaplastic thyroid cancer: the putative upstream role of SOX2." Thyroid**23**(7): 829-837.

BACKGROUND: Anaplastic thyroid carcinoma (ATC) is a rare and aggressive endocrine tumor with highly undifferentiated morphology. It has been suggested that cancer stem cells (CSCs) might play a central role in ATC. The objectives of this study were (i) to characterize CSCs from ex vivo ATC specimens by investigating the expression of several pluripotent stem cell markers, and (ii) to evaluate in vitro drug resistance modifications after specific CSC transcription factor switch-off. METHODS: In ex vivo experiments, eight formalin-fixed, paraffin-embedded ATC specimens were analyzed by reverse-transcription and real-time quantitative PCR and immunohistochemistry. In in vitro experiments using ATC SW1736 cells, the expression levels of OCT-4, NANOG, and ABCG2 and the sensitivity to either cisplatin or doxorubicin were evaluated after silencing. RESULTS: OCT-4, KLF4, and SOX2 transcription factors and C-KIT and THY-1 stem surface antigens showed variable up-regulation in all ATC cases. The SW1736 cell line was characterized by a high percentage of stem population (10.4+/-2.1% of cells were aldehyde dehydrogenase positive) and high expression of several CSC markers (SOX2, OCT4, NANOG, C-MYC, and SSEA4). SOX2 silencing

down-regulated OCT-4, NANOG, and ABCG2. SOX2 silencing sensitized SW1736 cells, causing a significant cell death increase (1.8-fold) in comparison to control cells with 10 muM cisplatin (93.9+/-3.4% vs. 52.6+/-9.4%, p<0.01) and 2.7 fold with 0.5 muM doxorubicin (45.8+/-9.9% vs. 17.1+/-3.4% p<0.01). ABCG2 silencing caused increased cell death with both cisplatin (74.9+/-1.4%) and doxorubicin treatment (74.1+/-0.1%) vs. no-target-treated cells (respectively, 45.8+/-1.0% and 48.6+/-1.0%, p<0.001). CONCLUSIONS: The characterization of CSCs in ATC through the analysis of multiple pluripotent stem cell markers might be useful in identifying cells with a stem-like phenotype capable of resisting conventional chemotherapy. In addition, our data demonstrate that SOX2 switch-off through ABCG2 transporter down-regulation has a major role in overcoming CSC chemotherapy resistance.

Carrasco-Garcia, E., et al. (2016)."Paradoxical role of SOX2 in gastric cancer."Am J Cancer Res**6**(4): 701-713.

Sox2 is a critical regulator of embryogenesis and necessary for cellular reprogramming. It also plays an important role in tissue homeostasis and regeneration, maintaining the population of undifferentiated adult stem cells. Like various developmental and stem cell genes, SOX2 is aberrantly expressed and amplified in several human cancers. Moreover, functional studies have shown that it regulates many biological processes including cell proliferation, apoptosis, self-renewal and invasion. While it is oncogenic in most cancers, SOX2 activity is controversial in gastric cancer, where it might behave as a tumor suppressor in some situations. In this review, we discuss its role in cancer biology, with particular attention to what is known about the involvement of SOX2 in gastric cancer biology.

Chen, J., et al. (2017). "CD59 Regulation by SOX2 Is Required for Epithelial Cancer Stem Cells to Evade Complement Surveillance." Stem Cell Reports**8**(1): 140-151.

Cancer stem cells (CSCs) are highly associated with therapy resistance and metastasis. Interplay between CSCs and various immune components is required for tumor survival. However, the response of CSCs to complement surveillance remains unknown. Herein, using stem-like sphere-forming cells prepared from a mammary tumor and a lung adenocarcinoma cell line, we found that CD59 was upregulated to protect CSCs from complement-dependent cytotoxicity. CD59 silencing significantly enhanced complement destruction and completely suppressed tumorigenesis in CSC-xenografted nude mice. Furthermore, we identified that SOX2 upregulates CD59 in epithelial CSCs. In addition, we revealed that SOX2 regulates the transcription of mCd59b, leading to selective mCD59b abundance in murine testis spermatogonial stem cells. Therefore, we demonstrated that CD59 regulation by SOX2 is required for stem cell evasion of complement surveillance. This finding highlights the importance of complement surveillance in eliminating CSCs and may suggest CD59 as a potential target for cancer therapy.

Chen, J., et al. (2018). "Downregulation of miR200c3p contributes to the resistance of breast cancer cells to paclitaxel by targeting SOX2." Oncol Rep**40**(6): 3821-3829.

Acquisition of resistance to paclitaxel is a major obstacle to successful treatment of breast cancer patients, but the molecular mechanisms underlying the development of drug resistance remain largely unclear. The aim of the present study was to investigate the role and mechanism of action of miR200c3p in the resistance of breast cancer to paclitaxel. It was observed that miR200c3p expression, as determined by reverse transcriptionquantitative polymerase chain reaction analysis, was significantly downregulated in paclitaxelresistant MCF7/Tax cells compared with parental MCF7 cells. Overexpression of miR200c3p increased the chemosensitivity to paclitaxel and enhanced apoptosis in MCF7/Tax cells, whereas the downregulation of miR200c3p exerted the opposite effect. In addition, upregulation of miR200c3p in MCF7/Tax cells suppressed the expression of sexdetermining region Ybox 2 (SOX2) at the mRNA and protein levels. Dualluciferase reporter assay demonstrated that SOX2 is a target of miR200c3p in MCF7/Tax cells. Moreover, knockdown of SOX2 expression increased chemosensitivity to paclitaxel and upregulated miR200c3p expression in MCF7/Tax cells. Taken together, the results of the present study indicated that miR200c3p plays a key role in the development of paclitaxel resistance in breast cancer, possibly partially through regulating SOX2 expression, suggesting that the miR200c3pSOX2 loop may serve as a potential target for the reversal of paclitaxel resistance in breast cancer.

Chen, S., et al. (2018). "Cancer-associated fibroblasts suppress SOX2-induced dysplasia in a lung squamous cancer coculture." Proc Natl Acad Sci U S A**115**(50): E11671-E11680.

Tumorigenesis depends on intricate interactions between genetically altered tumor cells and their surrounding microenvironment. While oncogenic drivers in lung squamous carcinoma (LUSC) have been described, the role of stroma in modulating tissue architecture, particularly cell polarity, remains unclear. Here, we report the establishment of a 3D coculture

system of LUSC epithelial cells with cancer-associated fibroblasts (CAFs) and extracellular matrix that together capture key components of the tumor microenvironment (TME). Single LUSC epithelial cells develop into acinar-like structures with 0.02% efficiency, and addition of CAFs provides proper tumor-stromal interactions within an appropriate 3D architectural context. Using this model, we recapitulate key pathological changes during tumorigenesis, from hyperplasia to dysplasia and eventually invasion, in malignant LUSC spheroids that undergo phenotypic switching in response to cell intrinsic and extrinsic changes. Overexpression of SOX2 is sufficient to mediate the transition from hyperplasia to dysplasia in LUSC spheroids, while the presence of CAFs makes them invasive. Unexpectedly, CAFs suppress the activity of high SOX2 levels, restore hyperplasia, and enhance the formation of acinar-like structures. Taken together, these observations suggest that stromal factors can override cell intrinsic oncogenic changes in determining the disease phenotype, thus providing fundamental evidence for the existence of dynamic reciprocity between the nucleus and the TME of LUSC.

Chen, S., et al. (2014). "SOX2 regulates apoptosis through MAP4K4-survivin signaling pathway in human lung cancer cells." Carcinogenesis**35**(3): 613-623.

Previous studies have implicated cancer stem cells in tumor recurrence and revealed that the stem cell gene SOX2 plays an important role in the tumor cell resistance to apoptosis. Nonetheless, the mechanism by which SOX2 regulates apoptosis signals remained undefined. Here, we demonstrated the surprising finding that silencing of the SOX2 gene effectively induces apoptosis via the activation of death receptor and mitochondrial signaling pathways in human non-small cell lung cancer cells. Unexpectedly, reverse transcription-PCR analysis suggested that downregulation of SOX2 leads to activation of MAP4K4, previously implicated in cell survival. Evaluation of the apoptotic pathways revealed an increased expression of key inducers of apoptosis, including tumor necrosis factor-alpha and p53, with concurrent attenuation of Survivin. Although p53 appeared dispensable for this pathway, the loss of Survivin in SOX2-deficient cells appeared critical for the observed MAP4K4 induced cell death. Rescue experiments revealed that SOX2-silencing-mediated killing was blocked by ectopic expression of Survivin, or by reduction of MAP4K4 expression. Clinically, expressions of Survivin and SOX2 were highly correlated with each other. The results reveal a key target of SOX2 expression and highlight the unexpected context-dependent role for MAP4K4, a pluripotent activator of several mitogen-activated protein kinase pathways, in regulating tumor cell survival.

Chhabra, R. (2018). "let-7i-5p, miR-181a-2-3p and EGF/PI3K/SOX2 axis coordinate to maintain cancer stem cell population in cervical cancer." Sci Rep**8**(1): 7840.

The characteristics of cancer stem cells (CSCs) and the genes responsible for their maintenance are highly variable in different cancers. Here, we identify the coordination among miRNAs and EGF pathway genes which is critical for the maintenance of CSCs in cervical cancer. The transcript analysis of CSCs enriched from cervical cancer cell lines (CaSki and HeLa) revealed a significant upregulation of SOX2. Since EGF receptor is frequently over expressed in cervical cancer, we hypothesized that EGF pathway may be responsible for the upregulation of SOX2. Also, the media used for CSC enrichment was supplemented with EGF. The hypothesis was validated as inhibiting the EGF/PI3K pathway suppressed the expression of SOX2 and reduced the CSC population. In addition, miRNA profiling identified miR-181a-2-3p and let-7i-5p as markedly reduced in CSCs. The exogenous expression of either of these miRNAs in CaSki cells inhibited the expression of SOX2 and subsequently reduced CSC population. In conclusion, this study highlights for the first time the contrasting role of let-7i-5p/ miR-181a-2-3p and EGF/PI3K/SOX2 axis in maintaining cervical CSCs. While the EGF pathway promotes CSC formation in cervical cancer by inducing SOX2, miR-181a-2-3p/let-7i-5p counteracts the EGF pathway by inhibiting SOX2, thereby reducing the CSC population.

Cho, Y. Y., et al. (2013). "Autophagy and cellular senescence mediated by Sox2 suppress malignancy of cancer cells." PLoS One**8**(2): e57172.

Autophagy is a critical cellular process required for maintaining cellular homeostasis in health and disease states, but the molecular mechanisms and impact of autophagy on cancer is not fully understood. Here, we found that Sox2, a key transcription factor in the regulation of the "stemness" of embryonic stem cells and induced-pluripotent stem cells, strongly induced autophagic phenomena, including intracellular vacuole formation and lysosomal activation in colon cancer cells. The activation occurred through Sox2-mediated ATG10 gene expression and resulted in the inhibition of cell proliferation and anchorage-independent colony growth ex vivo and tumor growth in vivo. Further, we found that Sox2-induced-autophagy enhanced cellular senescence by up-regulating tumor suppressors or senescence factors,

including p16(INK4a), p21 and phosphorylated p53 (Ser15). Notably, knockdown of ATG10 in Sox2-expressing colon cancer cells restored cancer cell properties. Taken together, our results demonstrated that regulation of autophagy mediated by Sox2 is a mechanism-driven novel strategy to treat human colon cancers.

Chou, M. Y., et al. (2015). "Sox2 expression involvement in the oncogenicity and radiochemoresistance of oral cancer stem cells."Oral Oncol**51**(1): 31-39.

OBJECTIVES: Sox2, a high-mobility-group DNA binding protein, is part of the key set of transcription factors that are involved in the maintenance of pluripotency and self-renewal in undifferentiated stem cells. A recent study has further suggested cancer stem cells (CSCs) are key contributors to radiochemoresistance and are responsible for oral squamous cell carcinoma (OSCC) progression. The aim of this study was to determine the emerging role of Sox2 in radiochemosensitivity of oral CSCs. METHODS: We determined the function of Sox2 on oncogenicity and radiochemosensitivity of OSCC by overexpression or silencing Sox2 in vitro and in vivo. RESULTS: Initially, Sox2 expression was increased in OSCC cell lines and OSCC specimens. Upregulated Sox2 is correlated with poor survival outcome of OSCC patients. Overexpression of Sox2 was demonstrated to enhance invasiveness, anchorage-independent growth, xenotransplantation tumourigenicity in OSCC cells. Targeting Sox2 to spheroid cells (SC) and ALDH1+CD44+ cells from OSCC significantly inhibited their CSCs and tumorigenic abilities. Down regulation of SOX2 in OSCC-SC was found to repress invasiveness and diminish epithelail-mesenchymal transition (EMT) traits. Furthermore, silencing Sox2 effectively suppressed the expression of drug-resistance and anti-apoptotic genes and increased the sensitivity of the cells to radiation combined cisplatin treatment. Finally, the in vivo therapeutic efficacy of targeting Sox2 synergistically suppressed tumorigenesis and improved the survival rate when used in combination with radiotherapy and cisplatin in OSCC-SC-transplanted immunocompromised mice. CONCLUSION: Sox2-mediated CSCs property is associated with the regulation of EMT and Sox2 s as therapeutic target in OSCC.

Chou, Y. T., et al. (2013). "The emerging role of SOX2 in cell proliferation and survival and its crosstalk with oncogenic signaling in lung cancer."Stem Cells**31**(12): 2607-2619.

Tumor cells have long been observed to share several biological characteristics with normal stem/progenitor cells; however, the oncogenic mechanisms underlying the lung stem/progenitor cell signaling remain elusive. Here, we report that SOX2, a self-renewal factor in lung stem/progenitor cells, is highly expressed in a subclass of lung cancer cells, the proliferation, survival, and chemoresistance of which are dependent on SOX2 signaling. Overexpression of SOX2 promotes oncogenic phenotypes in lung cancer cells; knockdown of SOX2 attenuated cell proliferation. We observed that SOX2 increased the expression of epidermal growth factor receptor (EGFR), and EGFR activation further upregulated SOX2 levels, forming a positive feedback loop. SOX2 expression promoted chemoresistance, and silencing of SOX2 perturbed mitochondrial function, causing marked apoptosis and autophagy. SOX2 induced BCL2L1, the ectopic expression of which rescued the effects of SOX2 silencing on apoptosis, autophagy, and mitochondrial function. SOX2 promoted tumor formation, along with increased cell proliferation in a xenograft mouse model. SOX2 expression is associated with poor prognosis in lung cancer patients; moreover, SOX2, EGFR, and BCL2L1 expression levels were significantly correlated in lung tumors. Our findings support the emerging role of SOX2 in cell proliferation and survival by eliciting oncogenic EGFR and BCL2L1 signaling with potential applications as a prognosis marker and a therapeutic target in lung cancer.

Corominas-Faja, B., et al. (2013). "Nuclear reprogramming of luminal-like breast cancer cells generates Sox2-overexpressing cancer stem-like cellular states harboring transcriptional activation of the mTOR pathway." Cell Cycle**12**(18): 3109-3124.

Energy metabolism plasticity enables stemness programs during the reprogramming of somatic cells to an induced pluripotent stem cell (iPSC) state. This relationship may introduce a new era in the understanding of Warburg's theory on the metabolic origin of cancer at the level of cancer stem cells (CSCs). Here, we used Yamanaka's stem cell technology in an attempt to create stable CSC research lines in which to dissect the transcriptional control of mTOR--the master switch of cellular catabolism and anabolism--in CSC-like states. The rare colonies with iPSC-like morphology, obtained following the viral transduction of the Oct4, Sox2, Klf4, and c-Myc (OSKM) stemness factors into MCF-7 luminal-like breast cancer cells (MCF-7/Rep), demonstrated an intermediate state between cancer cells and bona fide iPSCs. MCF-7/Rep cells notably overexpressed SOX2 and stage-specific embryonic antigen (SSEA)-4 proteins; however, other stemness-related markers (OCT4, NANOG, SSEA-1, TRA-1-60, and TRA-1-81) were found at low to moderate levels. The

transcriptional analyses of OSKM factors confirmed the strong but unique reactivation of the endogenous Sox2 stemness gene accompanied by the silencing of the exogenous Sox2 transgene in MCF-7/Rep cells. Some but not all MCF-7/Rep cells acquired strong alkaline phosphatase (AP) activity compared with MCF-7 parental cells. SOX2-overexpressing MCF-7/Rep cells contained drastically higher percentages of CD44(+) and ALDEFLUOR-stained ALDH (bright) cells than MCF-7 parental cells. The overlap between differentially expressed mTOR signaling-related genes in 3 different SOX2-overexpressing CSC-like cell lines revealed a notable downregulation of 3 genes, PRKAA1 (which codes for the catalytic alpha 1 subunit of AMPK), DDIT4/REDD1 (a stress response gene that operates as a negative regulator of mTOR), and DEPTOR (a naturally occurring endogenous inhibitor of mTOR activity). The insulin-receptor gene (INSR) was differentially upregulated in MCF-7/Rep cells. Consistent with the downregulation of AMPK expression, immunoblotting procedures confirmed upregulation of p70S6K and increased phosphorylation of mTOR in Sox2-overexpressing CSC-like cell populations. Using an in vitro model of the de novo generation of CSC-like states through the nuclear reprogramming of an established breast cancer cell line, we reveal that the transcriptional suppression of mTOR repressors is an intrinsic process occurring during the acquisition of CSC-like properties by differentiated populations of luminal-like breast cancer cells. This approach may provide a new path for obtaining information about preventing the appearance of CSCs through the modulation of the AMPK/mTOR pathway.

Correia, L. L., et al. (2017). "SOX2 Drives Bronchial Dysplasia in a Novel Organotypic Model of Early Human Squamous Lung Cancer." Am J Respir Crit Care Med**195**(11): 1494-1508.

RATIONALE: Improving the early detection and chemoprevention of lung cancer are key to improving outcomes. The pathobiology of early squamous lung cancer is poorly understood. We have shown that amplification of sex-determining region Y-box 2 (SOX2) is an early and consistent event in the pathogenesis of this disease, but its functional oncogenic potential remains uncertain. We tested the impact of deregulated SOX2 expression in a novel organotypic system that recreates the molecular and microenvironmental context in which squamous carcinogenesis occurs. OBJECTIVES: (1) To develop an in vitro model of bronchial dysplasia that recapitulates key molecular and phenotypic characteristics of the human disease; (2) to test the hypothesis that SOX2 deregulation is a key early event in the pathogenesis of bronchial dysplasia; and (3) to use the model for studies on pathogenesis and chemoprevention. METHODS: We engineered the inducible activation of oncogenes in immortalized bronchial epithelial cells. We used three-dimensional tissue culture to build an organotypic model of bronchial dysplasia. MEASUREMENTS AND MAIN RESULTS: We recapitulated human bronchial dysplasia in vitro. SOX2 deregulation drives dysplasia, and loss of tumor promoter 53 is a cooperating genetic event that potentiates the dysplastic phenotype. Deregulated SOX2 alters critical genes implicated in hallmarks of cancer progression. Targeted inhibition of AKT prevents the initiation of the dysplastic phenotype. CONCLUSIONS: In the appropriate genetic and microenvironmental context, acute deregulation of SOX2 drives bronchial dysplasia. This confirms its oncogenic potential in human cells and affords novel insights into the impact of SOX2 deregulation. This model can be used to test therapeutic agents aimed at chemoprevention.

Cox, J. L., et al. (2013). "The SOX2-interactome in brain cancer cells identifies the requirement of MSI2 and USP9X for the growth of brain tumor cells." PLoS One**8**(5): e62857.

Medulloblastomas and glioblastomas, the most common primary brain tumors in children and adults, respectively, are extremely difficult to treat. Efforts to identify novel proteins essential for the growth of these tumors may help to further our understanding of the biology of these tumors, as well as, identify targets for future therapies. The recent identification of multiple transcription factor-centric protein interaction landscapes in embryonic stem cells has identified numerous understudied proteins that are essential for the self-renewal of these stem cells. To identify novel proteins essential for the fate of brain tumor cells, we examined the protein interaction network of the transcription factor, SOX2, in medulloblastoma cells. For this purpose, Multidimensional Protein Identification Technology (MudPIT) identified >280 SOX2-associated proteins in the medulloblastoma cell line DAOY. To begin to understand the roles of SOX2-associated proteins in brain cancer, we focused on two SOX2-associated proteins, Musashi 2 (MSI2) and Ubiquitin Specific Protease 9x (USP9X). Recent studies have implicated MSI2, a putative RNA binding protein, and USP9X, a deubiquitinating enzyme, in several cancers, but not brain tumors. We demonstrate that knockdown of MSI2 significantly reduces the growth of DAOY cells as well as U87 and U118 glioblastoma cells. We also demonstrate that the knockdown of USP9X in DAOY, U87 and U118 brain tumor cells strongly reduces their growth. Together, our studies identify a large set of SOX2-associated proteins in DAOY medulloblastoma cells and identify

two proteins, MSI2 and USP9X, that warrant further investigation to determine whether they are potential therapeutic targets for brain cancer.

Das, T., et al. (2017)."Actinomycin D Down-regulates SOX2 Expression and Induces Death in Breast Cancer Stem Cells."Anticancer Res**37**(4): 1655-1663.

BACKGROUND/AIM: One of the major hurdles in the treatment of breast cancers is the inability of anti-cancer drugs to eliminate the breast cancer stem cells (BCSCs) population, which leads to disease relapse. The dearth in anti-cancer drugs that target BCSCs can be attributed to the absence of in vitro screening models that can not only recapitulate the tumor microenvironment consisting of BCSCs but also preserve the 3-dimensional (3D) architecture of in vivo tumors. MATERIALS AND METHODS: In our present study, we have developed a 3D cell culture system that shows: (i) enrichment of BCSCs, (ii) increased drug resistance, and (iii) generation of hypoxic conditions similar to tumors. RESULTS: Using this model, we were able to screen a FDA-approved diversity set and identify as well as validate actinomycin D as a potential anti-breast cancer agent. Interestingly, we show that actinomycin D specifically targets and down-regulates the expression of the stem cell transcription factor, Sox-2. Additionally, down-regulation of Sox-2 leads to depletion of the stem-cell population resulting in the inability of breast cancer cells to initiate tumor progression. CONCLUSION: This study demonstrates the utility of an in vivo-like 3D cell culture system for the identification and validation of anti-cancer agents that will have a better probability of success in the clinic.

Dhodapkar, K. M., et al. (2013). "SOX2-specific adaptive immunity and response to immunotherapy in non-small cell lung cancer." Oncoimmunology**2**(7): e25205.

Immunotherapeutic strategies including the blockade of programmed death 1 (PD-1) receptors hold promise for the treatment of various cancers including non-small cell lung carcinoma (NSCLC). Preclinical data suggest that pre-existing tumor immunity is important for disease regression upon checkpoint blockade-based therapies. However, the nature of antigen-specific T-cell responses that correlate with the clinical response to immunotherapy in NSCLC patients is not known. The embryonic stem cell gene SRY (sex determining region Y)-box 2 (SOX2) has recently emerged as a major oncogenic driver in NSCLC. Here, we show that nearly 50% of a cohort of NSCLC patients mounted both CD4(+) and CD8(+) T-cell responses against SOX2, which could be readily detected among peripheral blood mononuclear cells. T-cell responses against SOX2 were associated with NSCLC regression upon immunotherapy with anti-PD-1 monoclonal antibodies, whereas none of the patients lacking SOX2-specific T cells experienced disease regression following immune checkpoint blockade. Conversely, cellular and humoral responses against viral antigens or another tumor-associated antigen (NY-ESO-1) failed to correlate with the clinical response of NSCLC patients to immunotherapy. Of note, the administration of PD-1-blocking antibodies was associated with intramolecular epitope spread as well as with the amplification of SOX2-specific immune responses in vivo. These findings identify SOX2 as an important tumor-associated antigen in NSCLC and link the presence of SOX2-specific T cells with the clinical response of lung cancer patients to immunotherapy.

Dogan, I., et al. (2014). "SOX2 expression is an early event in a murine model of EGFR mutant lung cancer and promotes proliferation of a subset of EGFR mutant lung adenocarcinoma cell lines." Lung Cancer**85**(1): 1-6.

OBJECTIVES: Primary and acquired resistance to EGFR TKIs in EGFR mutant lung cancer occurs primarily through secondary mutations in EGFR or Met amplification. Drug resistance can also be mediated by expression of pluripotency transcription factors, such as OCT4, SOX2 and NANOG that decrease terminal differentiation. In this study, we investigated the expression and role of SOX2 in model systems of EGFR mutant tumors. MATERIALS AND METHODS: Immunoblotting or immunohistochemistry was used to assess expression of pluripotency transcription factors in lungs of transgenic mice or in human NSCLC cell lines. Expression of SOX2 was reduced by shRNA knockdown, and response to erlotinib and cellular proliferation were assessed. RESULTS AND CONCLUSION: Induction of mutant EGFR in transgenic CCSP-rtTA/TetO-EGFR (L858R/T790M) mice correlated with increased OCT4 and SOX2 expression in lung tissue prior to tumor development. Established lung tumors retained SOX2 expression. To assess a role for SOX2 in tumorigenesis, a panel of NSCLC cell lines with activating EGFR mutations was assessed for SOX2 expression. Two of six cell lines with mutant EGFR showed detectable SOX2 levels, suggesting SOX2 expression did not correlate with EGFR mutation status. To assess the role of SOX2 in these cell lines, HCC827 and H1975 cells were infected with lentivirus containing SOX2 shRNA. Knockdown of SOX2 decreased proliferation in both cell lines and increased sensitivity to erlotinib in HCC827 cells. Because constitutive activation of the PI3K/Akt pathway is associated with EGFR TKI

resistance, cells were treated with PI3K/AKT inhibitors and expression of SOX2 was examined. PI3K/Akt inhibitors decreased SOX2 expression in a time-dependent manner. These data suggest targeting SOX2 may provide therapeutic benefit in the subset of EGFR-mutant tumors with high constitutive levels of SOX2, and that until more direct means of inhibiting SOX2 are developed, PI3K/Akt inhibitors might be useful to inhibit SOX2 in EGFR TKI resistant tumors.

Dong, Z., et al. (2014). "Prognostic significance of SOX2 in head and neck cancer: a meta-analysis." Int J Clin Exp Med**7**(12): 5010-5020.

Sex determining region Y-box 2 (SOX2) has been identified as a putative cancer stem cells (CSCs) marker in Head and Neck Cancers (HNC). However, the clinicopathological and prognostic significance of SOX2 in HNC patients remains controversial. We reviewed the literature by performing a meta-analysis based on the data from 7 studies (9 cohorts) to evaluate the association between SOX2 and clinicopathological/prognostic parameters in patients with HNC. Pooled hazard ratio (HR) or odds ratio (OR) with its 95% confidence interval (CI) was used as the effect size estimate. Our analysis results suggested that high SOX2 expression predicted unfavorable OS (HR: 1.54, 95% CI: 1.09-2.18) and DFS (HR: 1.54, 95% CI: 1.13-2.10) of patients with HNC. In addition, increased SOX2 was also significantly associated with high tumor grade (OR: 1.86, 95% CI: 1.06-3.28), advanced TNM stage (OR: 4.22, 95% CI: 2.62-6.80), lymph node metastasis (OR: 2.25, 95% CI: 1.50-3.35) and distant metastasis (OR: 1.99, 95% CI: 1.26-3.15). Our study suggested that SOX2 expression can be served as a candidate unfavorable prognostic biomarker for HNC patients, indicating that it might be a potential therapeutic target.

Du, J., et al. (2015). "Overexpression of Class III beta-tubulin, Sox2, and nuclear Survivin is predictive of taxane resistance in patients with stage III ovarian epithelial cancer." BMC Cancer**15**: 536.

BACKGROUND: Class III beta-tubulin, Sox2, and Survivin play important roles in tumor survival and proliferation. However, the association of these three factors with clinicopathological characteristics, chemoresistance, and survival in patients with ovarian cancer remains controversial. METHODS: We investigated the predictive value and correlation among the expression levels of Class III beta-tubulin, Sox2, and Survivin in 110 patients with stage III ovarian epithelial cancer, including 58 patients who received taxane-based chemotherapy and 52 patients who received non-taxane-based chemotherapy. Expression of these three factors was immunohistochemically examined in 110 ovarian tumor tissues obtained from patients before chemotherapy. RESULTS: The positive expression rates for Class III beta-tubulin, Sox2, and Survivin in ovarian tumor tissues were 59.09 %, 61.82 % and 52.73 %, respectively. The expression of nuclear Survivin and Class III beta-tubulin was consistent with that of Sox2 ($p = 0.005$ and 0.020, respectively). Positive expression of Class III beta-tubulin, Sox2, and nuclear Survivin was significantly associated with chemoresistance to taxane-based chemotherapy ($p = 0.006$, 0.007, and 0.009, respectively), but not to non-taxane-based chemotherapy. Additionally, overexpression of Class III beta-tubulin, Sox2, and nuclear Survivin predicted poor progression-free survival in patients receiving taxane-based chemotherapy ($p = 0.032$, 0.005, and 0.004, respectively). CONCLUSIONS: These findings suggest that overexpression of Class III beta-tubulin, Sox2, and nuclear Survivin might be predictive of taxane resistance and poor progression-free survival in patients with stage III ovarian epithelial cancer. Expression of these three factors may show positive correlations in these patients.

Favaro, R., et al. (2014). "Sox2 is required to maintain cancer stem cells in a mouse model of high-grade oligodendroglioma." Cancer Res**74**(6): 1833-1844.

The stem cell-determining transcription factor Sox2 is required for the maintenance of normal neural stem cells. In this study, we investigated the requirement for Sox2 in neural cancer stem-like cells using a conditional genetic deletion mutant in a mouse model of platelet-derived growth factor-induced malignant oligodendroglioma. Transplanting wild-type oligodendroglioma cells into the brain generated lethal tumors, but mice transplanted with Sox2-deleted cells remained free of tumors. Loss of the tumor-initiating ability of Sox2-deleted cells was reversed by lentiviral-mediated expression of Sox2. In cell culture, Sox2-deleted tumor cells were highly sensitive to differentiation stimuli, displaying impaired proliferation, increased cell death, and aberrant differentiation. Gene expression analysis revealed an early transcriptional response to Sox2 loss. The observed requirement of oligodendroglioma stem cells for Sox2 suggested its relevance as a target for therapy. In support of this possibility, an immunotherapeutic approach based on immunization of mice with SOX2 peptides delayed tumor development and prolonged survival. Taken together, our results showed that Sox2 is essential for tumor initiation by mouse oligodendroglioma cells, and they illustrated a Sox2-directed strategy of immunotherapy to eradicate tumor-initiating cells.

Finicelli, M., et al. (2014). "Expression of stemness genes in primary breast cancer tissues: the role of SOX2 as a prognostic marker for detection of early recurrence." Oncotarget**5**(20): 9678-9688.

The events leading to breast cancer (BC) progression or recurrence are not completely understood and new prognostic markers aiming at identifying high risk-patients and to develop suitable therapy are highly demanded. Experimental evidences found in cancer cells a deregulated expression of some genes involved in governance of stem cell properties and demonstrated a relationship between stemness genes overexpression and poorly differentiated BC subtypes. In the present study 140 primary invasive BC specimens were collected. The expression profiles of 13 genes belonging to the OCT3/SOX2/NANOG/KLF4 core circuitry by RT-PCR were analyzed and any correlation between their expression and the BC clinic-pathological features (CPfs) and prognosis was investigated. In our cohort (117 samples), NANOG, GDF3 and SOX2 significantly correlated with grade 2, Nodes negative status and higher KI67 proliferation index, respectively (p=0.019, p=0.029, p= 0.035). According to multivariate analysis, SOX2 expression resulted independently associated with increased risk of recurrence (HR= 2,99; p= p=0,004) as well as Nodes status (HR=2,44; p=0,009) and T-size >1 (HR=1,77; p=0,035). Our study provides further proof of the suitable use of stemness genes in BC management. Interestingly, a prognostic role of SOX2, which seems to be a suitable marker of early recurrence irrespective of other clinicopathological features.

Forno, I., et al. (2015). "Deregulation of MiR-34b/Sox2 Predicts Prostate Cancer Progression." PLoS One**10**(6): e0130060.

Most men diagnosed with prostate cancer will have an indolent and curable disease, whereas approximately 15% of these patients will rapidly progress to a castrate-resistant and metastatic stage with high morbidity and mortality. Therefore, the identification of molecular signature (s) that detect men at risk of progressing disease remains a pressing and still unmet need for these patients. Here, we used an integrated discovery platform combining prostate cancer cell lines, a Transgenic Adenocarcinoma of the Mouse Prostate (TRAMP) model and clinically-annotated human tissue samples to identify loss of expression of microRNA-34b as consistently associated with prostate cancer relapse. Mechanistically, this was associated with epigenetics silencing of the MIR34B/C locus and increased DNA copy number loss, selectively in androgen-dependent prostate cancer. In turn, loss of miR-34b resulted in downstream deregulation and overexpression of the "stemness" marker, Sox2. These findings identify loss of miR-34b as a robust biomarker for prostate cancer progression in androgen-sensitive tumors, and anticipate a potential role of progenitor/stem cell signaling in this stage of disease.

Gan, L., et al. (2018). "Extracellular matrix protein 1 promotes cell metastasis and glucose metabolism by inducing integrin beta4/FAK/SOX2/HIF-1alpha signaling pathway in gastric cancer." Oncogene**37**(6): 744-755.

Extracellular matrix protein 1 (ECM1) is related to strong invasiveness and poor prognosis in major malignancies, but the underlying mechanism remains unknown. Here we aimed to elucidate the function of ECM1 on cell metastasis and glucose metabolism in gastric cancer (GC). The level of ECM1 in sera and tissues of patient with GC were positively correlated with tumor invasion and recurrence. Genetic manipulation of ECM1 expression affected cell metastasis and glucose metabolism in GC cell lines. Enhanced ECM1 expression facilitated gene expression levels associated with epithelial-mesenchymal transition (EMT) and glucose metabolism. Interestingly, our results indicated that ECM1 directly interacted with integrin beta4 (ITGB4) and activated ITGB4/focal adhesion kinase (FAK)/glycogen synthase kinase 3beta signaling pathway, which further induced the expression of transcription factor SOX2. Aberrant expression of SOX2 altered gene expression of EMT factors and glucose metabolism enzymes. Furthermore, SOX2 enhanced hypoxia-inducible factor alpha (HIF-1alpha) promoter activity to regulate glucose metabolism. The micro-positron emission tomography/computed tomography imaging of xenograft model showed that ECM1 substantially increased (18)F-fluorodeoxyglucose uptake in xenograft tumors. Using in vivo mouse tail vein injection experiments, ECM1 was also found to increase in lung surface metastasis. These findings provide evidence that ECM1 regulates GC cell metastasis and glucose metabolism by inducing ITGB4/FAK/SOX2/HIF-1alpha signal pathway and have important implications for the development of therapeutic target to prevent tumor metastasis and recurrence.

Gayyed, M. F. and E. R. Tawfiek (2015). "Utility of SOX2 and Livin Co-Expression in the Prognosis of Bladder Cancer With Bilharzial and Non-Bilharzial Bladder Status." World J Oncol**6**(5): 446-455.

BACKGROUND: The aim of this study was to investigate the expression of SOX2, a key transcription factor and livin, an apoptotic inhibitor in bladder transitional cell carcinoma (TCC) and

squamous cell carcinoma (SCC). Moreover, their prognostic significance was assessed. METHODS: The expressions of SOX2 and livin in 82 TCC and 35 SCC cases were detected by immunohistochemistry. RESULTS: SOX2 and livin were over-expressed in tumor tissues as compared to the corresponding adjacent non-neoplastic tissues. SOX2 and livin expressions were significantly associated with high tumor grade (P = 0.002 and P = 0.007, respectively) and high tumor stage (P = 0.027 and P = 0.033, respectively). No significant correlation was found between tumor and other clinicopathological factors such as age, gender and schistosomal status. Univariate analysis revealed that TCC and SCC patients with high SOX2 or livin expressions were significantly related to overall survival (P < 0.001, P = 0.025 for TCC patients and P = 0.041, P = 0.021 for SCC patients, respectively). Multivariate survival analysis further demonstrated that SOX2 expression was an independent prognostic factor for TCC patients (P = 0.015). CONCLUSIONS: SOX2 and livin may contribute to the progression of bladder carcinoma. SOX2/livin pathway regulates cancer stem cell survival so it could be targeting as an effective therapeutic strategy for cancer treatment.

Gopal, K., et al. (2016). "Oxidative stress induces the acquisition of cancer stem-like phenotype in breast cancer detectable by using a Sox2 regulatory region-2 (SRR2) reporter." Oncotarget**7**(3): 3111-3127.

We have previously identified a novel intra-tumoral dichotomy in breast cancer based on the differential responsiveness to a Sox2 reporter (SRR2), with cells responsive to SRR2 (RR) being more stem-like than unresponsive cells (RU). Here, we report that RR cells derived from MCF7 and ZR751 displayed a higher tolerance to oxidative stress than their RU counterparts, supporting the concept that the RR phenotype correlates with cancer stemness. Sox2 is directly implicated in this differential H2O2 tolerance, since siRNA knockdown of Sox2 in RR cells leveled this difference. Interestingly, H2O2 converted a proportion of RU cells into RR cells, as evidenced by their expression of luciferase and GFP, markers of SRR2 activity. Compared to RU cells, converted RR cells showed a significant increase in mammosphere formation and tolerance to H2O2. Converted RR cells also adopted the biochemical features of RR cells, as evidenced by their substantial increase in Sox2-SRR2 binding and the expression of 3 signature genes of RR cells (CD133, GPR49 and MUC15). Lastly, the H2O2-induced RU/RR conversion was detectable in a SCID mouse xenograft model and primary tumor cells. To conclude, the H2O2-induced RU/RR conversion has provided a novel model to study the acquisition of cancer stemness and plasticity.

Guo, L., et al. (2018). "17beta-estradiol regulates the malignancy of cancer stem-like cells derived from the MCF7 cell line partially through Sox2." Oncol Lett**15**(3): 3790-3795.

As a major common malignant tumor in women, the malignant behavior of breast cancer, which includes tumorigenesis and metastasis, is associated with estrogen, particularly 17beta-estradiol (E2). With accumulating evidence demonstrating that cancer stem-like cells (CSCs) serve a function in the malignant behavior of breast cancer, including metastasis, recurrence and chemoresistance, the effects of E2 on the physiological processes of CSCs have been attracting more attention. In the present study, in order to investigate the effects of E2 on CSCs, CSCs from the MCF7 breast cancer cell line were isolated and treated with 1, 10 and 50 nM E2. Detection of cell proliferation following E2 treatment revealed that 10 nM E2 treatment inhibited cell proliferation, whereas 50 nM E2 treatment resulted in the induction of apoptosis on CSCs. In order to further investigate the effects of E2 treatment on migration, colony formation and the self-renewal capacity of CSCs in vitro, cells were treated with 1 and 10 nM E2. As expected, compared with mock group, the self-renewal capacity of the CSCs was slightly increased by 10 nM E2 treatment, while 1 nM exhibited no observable effect. E2 treatment demonstrated different effects on the proliferation, migration, colony formation and self-renewal capacity of CSCs in a dose-dependent manner.

Gupta, N., et al. (2018). "Phosphorylation of Sox2 at Threonine 116 is a Potential Marker to Identify a Subset of Breast Cancer Cells with High Tumorigenecity and Stem-Like Features." Cancers (Basel)**10**(2).

We have previously identified a novel phenotypic dichotomy in breast cancer (BC) based on the response to a SRR2 (Sox2 regulatory region 2) reporter, with reporter responsive (RR) cells being more tumorigenic/stem-like than reporter unresponsive (RU) cells. Since the expression level of Sox2 is comparable between the two cell subsets, we hypothesized that post-translational modifications of Sox2 contribute to their differential reporter response and phenotypic differences. By liquid chromatography-mass spectrometry, we found Sox2 to be phosphorylated in RR but not RU cells. Threonine 116 is an important phosphorylation site, since transfection of the T116A mutant into RR cells significantly decreased the SRR2 reporter luciferase activity and the RR-associated phenotype. Oxidative stress-induced conversion of RU into RR cells was accompanied by Sox2 phosphorylation at T116 and increased Sox2-DNA

binding. In a cohort of BC, we found significant correlations between the proportion of tumor cells immuno-reactive with anti-phosphorylated Sox2(T116) and a high tumor grade (p = 0.006), vascular invasion (p = 0.001) and estrogen receptor expression (p = 0.032). In conclusion, our data suggests that phosphorylation of Sox2(T116) contributes to the tumorigenic/stem-like features in RR cells. Detection of phospho-Sox2(T116) may be useful in identifying a small subset of tumor cells carrying stem-like/tumorigenic features in BC.

Han, Y. T., et al. (2016). "Physcion inhibits the metastatic potential of human colorectal cancer SW620 cells in vitro by suppressing the transcription factor SOX2." Acta Pharmacol Sin**37**(2): 264-275.

AIM: Physcion, an anthraquinone derivative, exhibits hepatoprotective, anti-inflammatory, anti-microbial and anti-cancer activities. In this study we examined whether and how physcion inhibited metastatic potential of human colorectal cancer cells in vitro. METHODS: Human colorectal cancer cell line SW620 was tested. Cell migration and invasion were assessed using a wound healing and Transwell assay, respectively. The expression levels of transcription factor SOX2 in the cells were modulated with shRNA targeting SOX2 and SOX2 overexpressing plasmid. The expression of target molecules involved in epithelial-mesenchymal transition (EMT) process and the signaling pathways was determined with Western blots or qRT-PCR. ROS levels were measured using DCF-DA. RESULTS: Physcion (2.5, 5 mol/L) did not affect the cell viability, but dose-dependently inhibited the cell adhesion, migration and invasion. Physcion also inhibited the EMT process in the cells, as evidenced by the increased epithelial marker E-cadherin expression, and by decreased expression of mesenchymal markers N-cadherin, vimentin, fibronectin and alpha-SMA, as well as transcriptional repressors Snail, Slug and Twist. Physcion suppressed the expression of SOX2, whereas overexpression of SOX2 abrogated the inhibition of physcion on metastatic behaviors. Physcion markedly increased ROS production and phosphorylation of AMPK and GSK3beta in the cells, whereas the AMPK inhibitor compound C or the ROS inhibitor NAC abolished the inhibition of physcion on metastatic behaviors. CONCLUSION: Physcion inhibits the metastatic potential of human colorectal cancer cells in vitro via activating ROS/AMPK/GSK3beta signaling pathways and suppressing SOX2.

Hellner, K., et al. (2016). "Premalignant SOX2 overexpression in the fallopian tubes of ovarian cancer patients: Discovery and validation studies." EBioMedicine**10**: 137-149.

Current screening methods for ovarian cancer can only detect advanced disease. Earlier detection has proved difficult because the molecular precursors involved in the natural history of the disease are unknown. To identify early driver mutations in ovarian cancer cells, we used dense whole genome sequencing of micrometastases and microscopic residual disease collected at three time points over three years from a single patient during treatment for high-grade serous ovarian cancer (HGSOC). The functional and clinical significance of the identified mutations was examined using a combination of population-based whole genome sequencing, targeted deep sequencing, multi-center analysis of protein expression, loss of function experiments in an in-vivo reporter assay and mammalian models, and gain of function experiments in primary cultured fallopian tube epithelial (FTE) cells. We identified frequent mutations involving a 40kb distal repressor region for the key stem cell differentiation gene SOX2. In the apparently normal FTE, the region was also mutated. This was associated with a profound increase in SOX2 expression (p<2(-16)), which was not found in patients without cancer (n=108). Importantly, we show that SOX2 overexpression in FTE is nearly ubiquitous in patients with HGSOCs (n=100), and common in BRCA1-BRCA2 mutation carriers (n=71) who underwent prophylactic salpingo-oophorectomy. We propose that the finding of SOX2 overexpression in FTE could be exploited to develop biomarkers for detecting disease at a premalignant stage, which would reduce mortality from this devastating disease.

Herreros-Villanueva, M., et al. (2013). "SOX2 promotes dedifferentiation and imparts stem cell-like features to pancreatic cancer cells." Oncogenesis**2**: e61.

SOX2 (Sex-determining region Y (SRY)-Box2) has important functions during embryonic development and is involved in cancer stem cell (CSC) maintenance, in which it impairs cell growth and tumorigenicity. However, the function of SOX2 in pancreatic cancer cells is unclear. The objective of this study was to analyze SOX2 expression in human pancreatic tumors and determine the role of SOX2 in pancreatic cancer cells regulating CSC properties. In this report, we show that SOX2 is not expressed in normal pancreatic acinar or ductal cells. However, ectopic expression of SOX2 is observed in 19.3% of human pancreatic tumors. SOX2 knockdown in pancreatic cancer cells results in cell growth inhibition via cell cycle arrest associated with p21(Cip1) and p27(Kip1) induction, whereas SOX2 overexpression promotes S-phase entry and cell proliferation associated with cyclin D3 induction. SOX2 expression is associated with increased levels of the pancreatic

CSC markers ALDH1, ESA and CD44. Importantly, we show that SOX2 is enriched in the ESA (+)/CD44(+) CSC population from two different patient samples. Moreover, we show that SOX2 directly binds to the Snail, Slug and Twist promoters, leading to a loss of E-Cadherin and ZO-1 expression. Taken together, our findings show that SOX2 is aberrantly expressed in pancreatic cancer and contributes to cell proliferation and stemness/dedifferentiation through the regulation of a set of genes controlling G1/S transition and epithelial-to-mesenchymal transition (EMT) phenotype, suggesting that targeting SOX2-positive cancer cells could be a promising therapeutic strategy.

Hutz, K., et al. (2014). "The stem cell factor SOX2 regulates the tumorigenic potential in human gastric cancer cells." Carcinogenesis**35**(4): 942-950.

Gastric cancer (GC) is still one of the most common causes of cancer-related death worldwide, which is mainly attributable to late diagnosis and poor treatment options. Infection with Helicobacter pylori, different environmental factors and genetic alterations are known to influence the risk of developing gastric tumors. However, the molecular mechanisms involved in gastric carcinogenesis are still not fully understood, making it difficult to design targeted therapeutic approaches. Aberrant expression of the specific gastric differentiation marker SOX2 has been observed in stomach cancer. However, the role of SOX2 in gastric tumors has not been well established to date. To elucidate the role of SOX2 in gastric tumorigenesis, SOX2 transcriptional activity was blocked in AZ-521 cells. Interestingly, inhibition of SOX2 reduced cell proliferation and migration, increased apoptosis and induced changes in cell cycle. Blocking of SOX2 also reduced the tumorigenic potential of AZ-521 cells in vivo. In addition, correlation of SOX2 expression and proliferation was observed in a subset of human gastric tumors. Finally, target genes of SOX2 were for the first time identified by RNA microarray in GC cells. Taken together, the results presented here indicate that SOX2 controls several aspects related to GC development and progression by regulating the expression of members of important signaling pathways. These findings could provide new therapeutic options for a subset of GCs exhibiting SOX2 deregulation.

Iglesias, J. M., et al. (2014). "The Activation of the Sox2 RR2 Pluripotency Transcriptional Reporter in Human Breast Cancer Cell Lines is Dynamic and Labels Cells with Higher Tumorigenic Potential." Front Oncol**4**: 308.

The striking similarity displayed at the mechanistic level between tumorigenesis and the generation of induced pluripotent stem cells and the fact that genes and pathways relevant for embryonic development are reactivated during tumor progression highlights the link between pluripotency and cancer. Based on these observations, we tested whether it is possible to use a pluripotency-associated transcriptional reporter, whose activation is driven by the SRR2 enhancer from the Sox2 gene promoter (named S4+ reporter), to isolate cancer stem cells (CSCs) from breast cancer cell lines. The S4+ pluripotency transcriptional reporter allows the isolation of cells with enhanced tumorigenic potential and its activation was switched on and off in the cell lines studied, reflecting a plastic cellular process. Microarray analysis comparing the populations in which the reporter construct is active versus inactive showed that positive cells expressed higher mRNA levels of cytokines (IL-8, IL-6, TNF) and genes (such as ATF3, SNAI2, and KLF6) previously related with the CSC phenotype in breast cancer.

Iida, H., et al. (2012). "Hypoxia induces CD133 expression in human lung cancer cells by up-regulation of OCT3/4 and SOX2." Int J Oncol**40**(1): 71-79.

CD133 has been recognized as a specific cell surface marker for cancer stem cells in various tumors, although its biological functions and transcriptional regulation remain unclear. We found that the CD133 expression level was up-regulated in the lung cancer cell lines N417, H358, and A549, when these cell lines were cultured under hypoxic conditions. Among the five promoters (P1-P5) of human CD133 gene loci, P1 promoter was most strongly associated with hypoxia-induced promoter activity of CD133 gene expression. The P1 promoter possesses several cis-regulatory elements, including RUNT, GATA, ETS, OCT, SRY, and CREB-binding sites. A series of deletion and base substitution mutants of the P1 promoter revealed that OCT- and SRY-binding sites are important for hypoxia-induced promoter activity. The chromatin immunoprecipitation assay further confirmed the direct binding of Octamer biding trans-cription factor 3/4 (OCT4) and/or SRY-box containing gene 2 (SOX2) to the P1 promoter region of CD133 gene loci. In addition, the enhancement of both OCT4 and SOX2 expression by the alpha subunit of hypoxia-inducible factors (HIF1alpha and HIF2alpha) was required for hypoxia-induced CD133 expression. Knockdown of OCT4 or SOX2 expression in N417 cells with stabilized HIF1alpha and/or HIF2alpha abolished CD133P1 activity, while ectopic OCT4 or SOX2 expression triggers CD133P1 activity in the absence of HIF1alpha or HIF2alpha. Thus, in the hypoxic conditions, OCT4 and SOX2, both of which are induced by HIF1alpha/HIF2alpha. promote CD133

expression in the lung cancer cells via their direct interaction with the P1 promoter.

Inoue, Y., et al. (2015). "Clinicopathological and Survival Analysis of Japanese Patients with Resected Non-Small-Cell Lung Cancer Harboring NKX2-1, SETDB1, MET, HER2, SOX2, FGFR1, or PIK3CA Gene Amplification." J Thorac Oncol**10**(11): 1590-1600.

INTRODUCTION: Gene amplification is an important genetic change in cancer cells. We investigated the prevalence, clinicopathological characteristics, and prognostic value of NKX2-1 (also known as TTF-1), SETDB1, MET, HER2, SOX2, FGFR1, and PIK3CA amplification in Japanese patients with non-small-cell lung cancer (NSCLC). METHODS: The copy numbers of the seven above-mentioned genes were assessed using fluorescence in situ hybridization in a tissue microarray containing 282 surgically resected NSCLC specimens (164 adenocarcinoma [AC], 99 squamous cell carcinoma [SCC], and 19 others). Clinicopathological information were obtained from the medical records. RESULTS: NKX2-1, SETDB1, MET, HER2, SOX2, FGFR1, and PIK3CA gene amplification were observed in 30 of 277 (10.8%), 16 of 280 (5.7%), 38 of 278 (13.7%), 8 of 270 (3.0%), 34 of 278 (12.2%), 18 of 282 (6.4%), and 53 of 278 (19.1%) cases, respectively. Coamplification was detected in 16 of 156 (10.3%) AC patients and 35 of 93 (37.6%) SCC patients ($p < 0.0001$). NKX2-1 amplification was significantly related to an AC histology ($p = 0.004$), whereas SOX2, FGFR1, and PIK3CA amplifications were related to a SCC histology ($p < 0.0001$). Within the ACs, NKX2-1 and SETDB1 amplifications were markers of a shorter survival period. A multivariate Cox proportional hazards model revealed that NKX2-1 amplification was an independent predictor of poor survival (hazard ratio, 2.938; 95% confidence interval, 1.434-6.022; $p = 0.003$). Coamplification had impact on patient outcome in AC but not in entire NSCLC and SCC. CONCLUSIONS: The amplification status differed among the histological types of NSCLC. NKX2-1 amplification was an independent and the most practically important predictor of a poor prognosis among Japanese patients with AC.

Jeon, H. M., et al. (2011). "ID4 imparts chemoresistance and cancer stemness to glioma cells by derepressing miR-9*-mediated suppression of SOX2." Cancer Res**71**(9): 3410-3421.

Glioma stem cells (GSC) possess tumor-initiating potential and are relatively resistant to conventional chemotherapy and irradiation. Thus, they are considered to be major drivers for glioma initiation, progression, and recurrence. However, the precise mechanism governing acquisition of their drug resistance remains to be elucidated. Our previous study has shown that inhibitor of differentiation 4 (ID4) dedifferentiates Ink4a/Arf (-/-) mouse astrocytes and human glioma cells to glioma stem-like cells (induced GSCs or iGSCs). In this article, we report that ID4-driven iGSCs exhibit chemoresistant behavior to anticancer drugs through activation of ATP-binding cassette (ABC) transporters. We found that ID4 enhanced SOX2 protein expression by suppressing microRNA-9* (miR-9*), which can repress SOX2 by targeting its 3'-untranslated region. Consequently, ID4-mediated SOX2 induction enhanced ABCC3 and ABCC6 expression through direct transcriptional regulation, indicating that ID4 regulates the chemoresistance of iGSCs by promoting SOX2-mediated induction of ABC transporters. Furthermore, we found that short hairpin RNA-mediated knockdown of SOX2 in ID4-driven iGSCs resulted in loss of cancer stemness. Moreover, ectopic expression of SOX2 could dedifferentiate Ink4a/Arf (-/-) astrocytes and glioma cells to iGSCs, indicating a crucial role of SOX2 in genesis and maintenance of GSCs. Finally, we found that the significance of the ID4-miR-9*-SOX2-ABCC3/ABCC6 regulatory pathway is recapitulated in GSCs derived from patients with glioma. Together, our results reveal a novel regulatory mechanism by which ID4-driven suppression of miR-9* induces SOX2, which imparts stemness potential and chemoresistance to glioma cells and GSCs.

Ji, J., et al. (2014). "[Effect of Sox2 on proliferation of cervical squamous cancer cell line SiHa]."Sichuan Da Xue Xue Bao Yi Xue Ban**45**(5): 785-788.

OBJECTIVE: To investigate the effect and its mechanism of stem cell related transcription factor Sox2 on the proliferation of cervical squamous carcinoma cell line SiHa. METHODS: Plasmid pIRES-EGFP-Sox2 or empty plasmid (pIRES-EGFP-empty) was stably transfected into SiHa cells. The expression of Sox2 was detected by both RT-PCT and Western blot. The effects of Sox2 on cellular proliferation and cell cycle were studied by MTT assay and flow cytometry (FCM) respectively. The expression of cell cycle related protein CyclinD1 was detected by Western blot. RESULTS: Compared to SiHa-EGFP cells, the expression of Sox2 was obviously up-regulated in SiHaSox2 cells ($P < 0.01$). MTT result showed that SiHa-Sox2 cells grew faster than the control cells. The over expression of Sox2 increased the proportion of transfected cells in phase S. The increased expression of CyclinD1 was further detected after the successful expression of Sox2 ($P < 0.05$). CONCLUSION: Sox2 could enhance the

proliferation of cervical squamous cancer cells in the manner of up-regulating CyclinD1 expression.

Jia, X., et al. (2011). "SOX2 promotes tumorigenesis and increases the anti-apoptotic property of human prostate cancer cell." J Mol Cell Biol**3**(4): 230-238.

SRY-related HMG-box gene 2 (SOX2) is one of the key regulatory genes that maintain the pluripotency and self-renewal properties in embryonic stem cells. Here we used immunohistochemistry to analyze the expression of SOX2 in human prostate tissues and found it contributed to tumorigenesis and correlated with histologic grade and Gleason score. We further investigated SOX2's function in cell growth and apoptosis process by using a human prostate cancer cell line DU145 with SOX2 overexpression or down-regulation. Cell cycle assay revealed that SOX2 promoted cell growth and increased the percentage of cells in S phase. In vitro and in vivo xenograft experiments in NOD/SCID mice further demonstrated that SOX2 increased the apoptosis-resistant properties of DU145 cells with decreased function of store-operated Ca (2+) entry and reduced expression of Orai1 at both mRNA and protein levels, suggesting a potential mechanism that contributes to the anti-apoptotic property of SOX2. To our knowledge, this study is the first to investigate SOX2's function in tumorigenesis and apoptosis of human prostate cancer and to elucidate its regulatory effect on the activity of store-operated Ca (2+) channels. Our results support the concept that SOX2 has the potential to be a significant marker to evaluate the progression of prostate cancer and serve as a potentially useful target for prostate cancer therapy.

Jia, Y., et al. (2018). "The role of GLI-SOX2 signaling axis for gemcitabine resistance in pancreatic cancer."Oncogene.

Pancreatic cancer, mostly pancreatic ductal adenocarcinomas (PDAC), is one of the most lethal cancers, with a dismal median survival around 8 months. PDAC is notoriously resistant to chemotherapy. Thus far, numerous attempts using novel targeted therapies and immunotherapies yielded limited clinical benefits for pancreatic cancer patients. It is hoped that delineating the molecular mechanisms underlying drug resistance in pancreatic cancer may provide novel therapeutic options. Using acquired gemcitabine resistant pancreatic cell lines, we revealed an important role of the GLI-SOX2 signaling axis for regulation of gemcitabine sensitivity in vitro and in animal models. Down-regulation of GLI transcriptional factors (GLI1 or GLI2), but not SMO signaling inhibition, reduces tumor sphere formation, a characteristics of tumor initiating cell (TIC). Down-regulation of GLI transcription factors also decreased expression of TIC marker CD24. Similarly, high SOX2 expression is associated with gemcitabine resistance whereas down-regulation of SOX2 sensitizes pancreatic cancer cells to gemcitabine treatment. We further revealed that elevated SOX2 expression is associated with an increase in GLI1 or GLI2 expression. Our ChIP assay revealed that GLI proteins are associated with a putative Gli binding site within the SOX2 promoter, suggesting a more direct regulation of SOX2 by GLI transcription factors. The relevance of our findings to human disease was revealed in human cancer specimens. We found that high SOX2 protein expression is associated with frequent tumor relapse and poor survival in stage II PDAC patients (all of them underwent gemcitabine treatment), indicating that reduced SOX2 expression or down-regulation of GLI transcription factors may be effective in sensitizing pancreatic cancer cells to gemcitabine treatment.

Jung, K., et al. (2014). "YB-1 regulates Sox2 to coordinately sustain stemness and tumorigenic properties in a phenotypically distinct subset of breast cancer cells." BMC Cancer**14**: 328.

BACKGROUND: Sox2, a transcription factor and an embryonic stem cell marker, has been implicated in the pathogenesis of breast cancer (BC). YB-1 is another transcription factor that has been shown to promote stemness in BC cells. METHODS: Western blotting, quantitative PCR, and siRNAs were used to query the regulatory relationships between YB-1, Sox2, and their downstream targets. Chromatin immunoprecipitation was used to detect YB-1 interactions at the Sox2 promoter. Mammosphere and soft agar assays were used to assess the phenotypic consequences of YB-1 knockdown. RESULTS: Here, we report that YB-1 regulates Sox2. YB-1 was found to bind to the SOX2 promoter and down-regulate its expression in MCF7 and ZR751. The regulatory interaction between YB-1 and Sox2 was drastically different between the two phenotypically distinct cell subsets, purified based on their differential response to a Sox2 reporter. They are referred to as the reporter unresponsive (RU) cells and the reporter responsive (RR) cells. Upon siRNA knockdown of YB-1, RU cells showed an increase in Sox2 expression but no change in Sox2 reporter activity; in contrast, RR cells exhibited increased expression and reporter activity of Sox2. Correlating with these findings, YB-1 knockdown induced a differential response in the expression of genes known to be regulated by both Sox2 and YB-1 (e.g. CCND1 and ITGA6). For instance, in response to YB-1 knockdown, CCND1 and ITGA6 expression were decreased or unchanged in RU cells but paradoxically increased in RR cells.

Compared to RU cells, RR cells were significantly more resistant to the suppression of mammosphere formation due to YB-1 knockdown. Importantly, mammospheres derived from parental MCF7 cells treated with YB-1 siRNA knockdown exhibited higher expression levels of SOX2 and its downstream targets. CONCLUSIONS: To conclude, in a subset of BC cells, namely RR cells, YB-1 regulates Sox2 to coordinately maintain stemness and tumorigenic properties.

Kar, S., et al. (2017). "SOX2 function and Hedgehog signaling pathway are co-conspirators in promoting androgen independent prostate cancer." Biochim Biophys Acta Mol Basis Dis**1863**(1): 253-265.

Developmentally inclined hedgehog (HH) signaling pathway and pluripotency inducing transcription factor SOX2 have been known to work syngerstically during cellular reprogramming events to facilitate efficient differentiation. Hence, it is not surprising that both the factors are actively involved in arbitrating malignant growth, including prostate cancer progression. Here, we have described in details the potential mechanisms by which SOX2 effects neoplastic characteristics in prostate cancer and investigated the consequences of simultaneous down-regulation of SOX2 and HH pathway in androgen-independent human prostate cancer cells. Expression of SOX2 has been determined by qRT-PCR, western blot, immunohistochemistry and immunocytochemistry analyses; its functional role determined by gene knockdown using RNAi and over-expression via chemical activation in HaCaT, DU145 and PC-3 cells. Changes in level of cell proliferation, migration and apoptosis profiles were measured by MTT, FACS, chromatin condensation and scratch assays respectively. SOX2 was expressed in all the three cell lines and its inhibition reduced cell proliferation and induced apoptosis. Most importantly, when both SOX2 and HH pathway were targeted simultaneously, cell proliferation was greatly reduced, apoptotic cell population increased drastically and migration potential was reduced. Moreover, gene expression of EMT markers such as E-cadherin and apoptosis related Bcl-2 and Bax was also investigated wherein decrease in E-cadherin and Bcl-2 levels and increase in Bax expression further substantiating our claim. These findings could provide the basis for a novel therapeutic strategy targeting both the effector i.e. SOX2 and perpetuator i.e. HH pathway of aggressive tumorigenic properties in androgen independent prostate cancer.

Karachaliou, N., et al. (2013). "The role of SOX2 in small cell lung cancer, lung adenocarcinoma and squamous cell carcinoma of the lung."Transl Lung Cancer Res**2**(3): 172-179.

SOX2 is a stem cell transcription factor that plays a crucial role in the regulation of embryonic development. It is one of the genes in a set of factors (Oct4, SOX2, Nanog) that are able to reprogram human somatic cells to pluripotent stem cells. Overexpression of SOX2 has been described in all types of lung cancer tissues, including small cell and squamous cell carcinoma but also adenocarcinoma. An in-depth view of the spectrum of genomic alterations in small cell lung cancer (SCLC) has identified SOX2 as a potential target for therapeutic intervention. Amplification of 3q, the most common genomic aberration in squamous lung cancer, has been demonstrated in the evolution of preinvasive squamous lung cancer and implicates SOX2 as a key target of this dynamic process, making SOX2 and its downstream effector components potential targets for biological therapeutics of squamous carcinomas. SOX2 is expressed in nearly 20% of lung adenocarcinoma and is associated with poor prognosis. SOX2 activity was found to promote squamous identity instead of a loss of cellular differentiation consistent with the role of SOX2 as a "lineage-survival oncogene." Interestingly, SOX2 transcription factor is the predominant downstream target of EGFR signaling and plays a major role in self-renewal growth and expansion of side population cells. In light of the complex actions of SOX2 in regulating normal and tumor development, the elucidation of SOX2-dependent pathways may identify new therapeutic vulnerabilities in lung cancer and uncover additional common pathways between cancer, normal development, and the maintenance of pluripotency.

Kaufhold, S., et al. (2016). "Yin Yang 1 is associated with cancer stem cell transcription factors (SOX2, OCT4, BMI1) and clinical implication." J Exp Clin Cancer Res**35**: 84.

The transcription factor Yin Yang 1 (YY1) is frequently overexpressed in cancerous tissues compared to normal tissues and has regulatory roles in cell proliferation, cell viability, epithelial-mesenchymal transition, metastasis and drug/immune resistance. YY1 shares many properties with cancer stem cells (CSCs) that drive tumorigenesis, metastasis and drug resistance and are regulated by overexpression of certain transcription factors, including SOX2, OCT4 (POU5F1), BMI1 and NANOG. Based on these similarities, it was expected that YY1 expression would be associated with SOX2, OCT4, BMI1, and NANOG's expressions and activities. Data mining from the proteomic tissue-based datasets from the Human Protein Atlas were used for protein expression patterns of YY1 and the

four CSC markers in 17 types of cancer, including both solid and hematological malignancies. A close association was revealed between the frequency of expressions of YY1 and SOX2 as well as SOX2 and OCT4 in all cancers analyzed. Two types of dynamics were identified based on the nature of their association, namely, inverse or direct, between YY1 and SOX2. These two dynamics define distinctive patterns of BMI1 and OCT4 expressions. The relationship between YY1 and SOX2 expressions as well as the expressions of BMI1 and OCT4 resulted in the classification of four groups of cancers with distinct molecular signatures: (1) Prostate, lung, cervical, endometrial, ovarian and glioma cancers (YY1(lo)SOX2(hi)BMI1(hi)OCT4(hi)) (2) Skin, testis and breast cancers (YY1(hi)SOX2(lo)BMI1(hi)OCT4(hi)) (3) Liver, stomach, renal, pancreatic and urothelial cancers (YY1(lo)SOX2(lo)BMI1(hi)OCT4(hi)) and (4) Colorectal cancer, lymphoma and melanoma (YY1(hi)SOX2(hi)BMI1(lo)OCT4(hi)). A regulatory loop is proposed consisting of the cross-talk between the NF-kB/PI3K/AKT pathways and the downstream inter-regulation of target gene products YY1, OCT4, SOX2 and BMI1.

Keysar, S. B., et al. (2017). "Regulation of Head and Neck Squamous Cancer Stem Cells by PI3K and SOX2."J Natl Cancer Inst**109**(1).

Background: We have an incomplete understanding of the differences between cancer stem cells (CSCs) in human papillomavirus-positive (HPV-positive) and -negative (HPV-negative) head and neck squamous cell cancer (HNSCC). The PI3K pathway has the most frequent activating genetic events in HNSCC (especially HPV-positive driven), but the differential signaling between CSCs and non-CSCs is also unknown. Methods: We addressed these unresolved questions using CSCs identified from 10 HNSCC patient-derived xenografts (PDXs). Sored populations were serially passaged in nude mice to evaluate tumorigenicity and tumor recapitulation. The transcription profile of HNSCC CSCs was characterized by mRNA sequencing, and the susceptibility of CSCs to therapy was investigated using an in vivo model. SOX2 transcriptional activity was used to follow the asymmetric division of PDX-derived CSCs. All statistical tests were two-sided. Results: CSCs were enriched by high aldehyde dehydrogenase (ALDH) activity and CD44 expression and were similar between HPV-positive and HPV-negative cases (percent tumor formation injecting </= 1x10(3) cells: ALDH (+)CD44(high) = 65.8%, ALDH (-)CD44(high) = 33.1%, ALDH (+)CD44(high) = 20.0%; and injecting 1x10(5) cells: ALDH (-)CD44(low) = 4.4%). CSCs were resistant to conventional therapy and had PI3K/mTOR pathway overexpression (GSEA pathway enrichment, P <.001), and PI3K inhibition in vivo decreased their tumorigenicity (40.0%-100.0% across cases). PI3K/mTOR directly regulated SOX2 protein levels, and SOX2 in turn activated ALDH1A1 (P <.001 013C and 067C) expression and ALDH activity (ALDH (+) [%] empty-control vs SOX2, 0.4% +/- 0.4% vs 14.5% +/- 9.8%, P =.03 for 013C and 1.7% +/- 1.3% vs 3.6% +/- 3.4%, P =.04 for 067C) in 013C and 067 cells. SOX2 enhanced sphere and tumor growth (spheres/well, 013C P <.001 and 067C P =.04) and therapy resistance. SOX2 expression prompted mesenchymal-to-epithelial transition (MET) by inducing CDH1 (013C P =.002, 067C P =.01), followed by asymmetric division and proliferation, which contributed to tumor formation. Conclusions: The molecular link between PI3K activation and CSC properties found in this study provides insights into therapeutic strategies for HNSCC. Constitutive expression of SOX2 in HNSCC cells generates a CSC-like population that enables CSC studies.

Kim, B. W., et al. (2015). "Clinical significance of OCT4 and SOX2 protein expression in cervical cancer."BMC Cancer**15**: 1015.

BACKGROUND: Cancer stem cell markers have become a major research focus because of their relationship with radiation or chemotherapy resistance in cancer therapy. Cancer stem cell markers including OCT4 and SOX2 have been found in various solid tumors. Here, we investigate the expression and clinical significance of OCT4 and SOX2 in cervical cancer. METHODS: To define the clinical significance of OCT4 and SOX2 expression, we performed immunohistochemistry for OCT4 and SOX2 on 305 normal cervical epithelium samples, 289 cervical intraepithelial neoplasia samples, and 161 cervical cancer cases and compared the data with clinicopathologic factors, including survival rates of patients with cervical cancer. RESULTS: OCT4 and SOX2 expression was higher in cervical cancer than normal cervix (both p < 0.001). OCT4 overexpression was associated with lymphovascular space invasion (p = 0.045), whereas loss of SOX2 expression was correlated with large tumor size (p = 0.015). Notably, OCT4 and SOX2 were significantly co-expressed in premalignant cervical lesions, but not in malignant cervical tumor. OCT4 overexpression showed worse 5-year disease-free and overall survival rates (p = 0.012 and p = 0.021, respectively) when compared to the low-expression group, while SOX2 expression showed favorable overall survival (p = 0.025). Cox regression analysis showed that OCT4 was an independent risk factor (hazard ratio = 11.23, 95 % CI, 1.31 - 95.6; p = 0.027) for overall survival while

SOX2 overexpression showed low hazard ratio for death (hazard ratio = 0.220, 95 % CI, 0.06-0.72; p = 0.013). CONCLUSIONS: These results suggest that OCT4 overexpression and loss of SOX2 expression are strongly associated with poor prognosis in patients with cervical cancer.

Kitamura, H., et al. (2013). "Prognostic impact of the expression of ALDH1 and SOX2 in urothelial cancer of the upper urinary tract."Mod Pathol**26**(1): 117-124.

Aldehyde dehydrogenase 1 (ALDH1) and sex determining region-Y-related high mobility group box 2 (SOX2) have been identified as putative cancer stem-like cell/tumor-initiating cell markers in various cancer tissues. The aim of this study was to elucidate the prognostic impact of these putative cancer stem-like cell/tumor-initiating cell markers in upper urinary tract urothelial cell carcinoma. Immunohistochemical staining for ALDH1 and SOX2 was carried out on archival specimens from 125 patients with upper urinary tract urothelial cell carcinoma who underwent radical nephroureterectomy. The prognostic value of ALDH1 and SOX2 expression and other clinicopathological features was evaluated. On univariate analysis, tumor grade, pathological T stage, pathological N stage, lymphovascular invasion, ALDH1 expression and SOX2 expression were associated with a poor prognosis. On multivariate analysis, the independent factors of prognosis were tumor grade (P=0.014), pathological N stage (P=0.005) and ALDH1 expression (P=0.002). In subgroup analysis, those subgroups with no positive, one positive or two positive results in immunohistochemistry for ALDH1 and SOX2 expression had estimated 5-year cancer-specific survival rates of 80%, 49% and 22%, respectively (P<0.001). Neither ALDH1 nor SOX2 expression correlated with intravesical recurrence after radical nephroureterectomy. These findings suggest that cancer stem-like cells/tumor-initiating cells are linked to more aggressive behavior of upper urinary tract urothelial cell carcinoma, supporting the current cancer stem cell hypothesis. Thus, therapeutic targeting of cancer stem-like cells/tumor-initiating cells in upper urinary tract urothelial cell carcinoma is a future possibility.

Kregel, S., et al. (2013). "Sox2 is an androgen receptor-repressed gene that promotes castration-resistant prostate cancer." PLoS One**8**(1): e53701.

Despite advances in detection and therapy, castration-resistant prostate cancer continues to be a major clinical problem. The aberrant activity of stem cell pathways, and their regulation by the Androgen Receptor (AR), has the potential to provide insight into novel mechanisms and pathways to prevent and treat advanced, castrate-resistant prostate cancers. To this end, we investigated the role of the embryonic stem cell regulator Sox2 [SRY (sex determining region Y)-box 2] in normal and malignant prostate epithelial cells. In the normal prostate, Sox2 is expressed in a portion of basal epithelial cells. Prostate tumors were either Sox2-positive or Sox2-negative, with the percentage of Sox2-positive tumors increasing with Gleason Score and metastases. In the castration-resistant prostate cancer cell line CWR-R1, endogenous expression of Sox2 was repressed by AR signaling, and AR chromatin-IP shows that AR binds the enhancer element within the Sox2 promoter. Likewise, in normal prostate epithelial cells and human embryonic stem cells, increased AR signaling also decreases Sox2 expression. Resistance to the anti-androgen MDV3100 results in a marked increase in Sox2 expression within three prostate cancer cell lines, and in the castration-sensitive LAPC-4 prostate cancer cell line ectopic expression of Sox2 was sufficient to promote castration-resistant tumor formation. Loss of Sox2 expression in the castration-resistant CWR-R1 prostate cancer cell line inhibited cell growth. Up-regulation of Sox2 was not associated with increased CD133 expression but was associated with increased FGF5 (Fibroblast Growth Factor 5) expression. These data propose a model of elevated Sox2 expression due to loss of AR-mediated repression during castration, and consequent castration-resistance via mechanisms not involving induction of canonical embryonic stem cell pathways.

Kuo, H. Y., et al. (2016). "Galectin-3 modulates the EGFR signalling-mediated regulation of Sox2 expression via c-Myc in lung cancer." Glycobiology**26**(2): 155-165.

Galectin-3 is a ubiquitous lectin exerting multiple cellular functions such as RNA splicing, protein trafficking and apoptosis. Its expression is positively correlated with the poor prognosis in lung cancer patients. Galectin-3 can promote cancer progression through its effects on cell proliferation, cell survival or cancer metastasis. However, the role of galectin-3 in the regulation of cancer stem-like cells (CSCs) is still unclear. Here, we investigated the hypothesis that galectin-3 might regulate lung CSCs via the EGF receptor (EGFR) signaling pathway. In our study, galectin-3 facilitated EGFR activation and enhanced the sphere formation activity of lung cancer cells. Furthermore, galectin-3 promoted Sox2 expression in an EGFR activation-dependent manner; importantly, forced expression of Sox2 blunted the effect of galectin-3 knockdown on lung cancer sphere formation ability. These results suggest that galectin-3 promotes EGFR activation leading to the upregulation

of Sox2 expression and lung CSCs properties. Moreover, we showed that the carbohydrate-binding activity of galectin-3 was important for the regulation of EGFR activation, Sox2 expression and sphere formation. We have recently reported that c-Myc is a transcriptional activator of Sox2. We further found that galectin-3 enhanced c-Myc protein stability leading to increased c-Myc binding to the Sox2 gene promoter. We also examined the effect of the stemness factors, Oct4, Nanog and Sox2 on the expression of galectin-3. We found that Oct4 enhanced galectin-3 expression. Our results together suggest that galectin-3 enhances lung cancer stemness through the EGFR/c-Myc/Sox2 axis; Oct4, in turn, promotes galectin-3 expression, forming a positive regulatory loop in lung CSCs.

Lee, S., et al. (2017). "Crosstalks between Raf-kinase inhibitor protein and cancer stem cell transcription factors (Oct4, KLF4, Sox2, Nanog)." Tumour Biol**39**(4): 1010428317692253.

Raf-kinase inhibitor protein has been reported to inhibit both the Raf/mitogen extracellular signal-regulated kinase/extracellular signal-regulated kinase and nuclear factor kappa-light-chain of activated B cells pathways. It has also been reported in cancers that Raf-kinase inhibitor protein behaves as a metastatic suppressor as well as a chemo-immunosensitizing factor to drug/immune-mediated apoptosis. The majority of cancers exhibit low or no levels of Raf-kinase inhibitor protein. Hence, the activities of Raf-kinase inhibitor protein contrast, in part, to those mediated by several cancer stem cell transcription factors for their roles in resistance and metastasis. In this review, the existence of crosstalks in the signaling pathways between Raf-kinase inhibitor protein and several cancer stem cell transcription factors (Oct4, KLF4, Sox2 and Nanog) was assembled. Oct4 is induced by Lin28, and Raf-kinase inhibitor protein inhibits the microRNA binding protein Lin28. The expression of Raf-kinase inhibitor protein inversely correlates with the expression of Oct4. KLF4 does not interact directly with Raf-kinase inhibitor protein, but rather interacts indirectly via Raf-kinase inhibitor protein's regulation of the Oct4/Sox2/KLF4 complex through the mitogen-activated protein kinase pathway. The mechanism by which Raf-kinase inhibitor protein inhibits Sox2 is via the inhibition of the mitogen-activated protein kinase pathway by Raf-kinase inhibitor protein. Thus, Raf-kinase inhibitor protein's relationship with Sox2 is via its regulation of Oct4. Inhibition of extracellular signal-regulated kinase by Raf-kinase inhibitor protein results in the upregulation of Nanog. The inhibition of Oct4 by Raf-kinase inhibitor protein results in the failure of the heterodimer formation of Oct4 and Sox2 that is necessary to bind to the Nanog promoter for the transcription of Nanog. The findings revealed that there exists a direct correlation between the expression of Raf-kinase inhibitor protein and the expression of each of the above transcription factors. Based on these analyses, we suggest that the expression level of Raf-kinase inhibitor protein may be involved in the regulation of the cancer stem cell phenotype.

Leis, O., et al. (2012)."Sox2 expression in breast tumours and activation in breast cancer stem cells."Oncogene**31**(11): 1354-1365.

The cancer stem cell (CSC) model does not imply that tumours are generated from transformed tissue stem cells. The target of transformation could be a tissue stem cell, a progenitor cell, or a differentiated cell that acquires self-renewal ability. The observation that induced pluripotency reprogramming and cancer are related has lead to the speculation that CSCs may arise through a reprogramming-like mechanism. Expression of pluripotency genes (Oct4, Nanog and Sox2) was tested in breast tumours by immunohistochemistry and it was found that Sox2 is expressed in early stage breast tumours. However, expression of Oct4 or Nanog was not found. Mammosphere formation in culture was used to reveal stem cell properties, where expression of Sox2, but not Oct4 or Nanog, was induced. Over-expression of Sox2 increased mammosphere formation, effect dependent on continuous Sox2 expression; furthermore, Sox2 knockdown prevented mammosphere formation and delayed tumour formation in xenograft tumour initiation models. Induction of Sox2 expression was achieved through activation of the distal enhancer of Sox2 promoter upon sphere formation, the same element that controls Sox2 transcription in pluripotent stem cells. These findings suggest that reactivation of Sox2 represents an early step in breast tumour initiation, explaining tumour heterogeneity by placing the tumour-initiating event in any cell along the axis of mammary differentiation.

Lundberg, I. V., et al. (2016). "SOX2 expression is associated with a cancer stem cell state and down-regulation of CDX2 in colorectal cancer." BMC Cancer**16**: 471.

BACKGROUND: To improve current treatment strategies for patients with aggressive colorectal cancer (CRC), the molecular understanding of subgroups of CRC with poor prognosis is of vast importance. SOX2 positive tumors have been associated with a poor patient outcome, but the functional role of SOX2 in CRC patient prognosis is still unclear. METHODS: An in vitro cell culture model expressing SOX2 was used to investigate the functional role of SOX2 in CRC. In vitro findings

were verified using RNA from fresh frozen tumor tissue or immunohistochemistry on formalin fixed paraffin embedded (FFPE) tumor tissue from a cohort of 445 CRC patients. RESULTS: Using our in vitro model, we found that SOX2 expressing cells displayed several characteristics of cancer stem cells; such as a decreased proliferative rate, a spheroid growth pattern, and increased expression of stem cell markers CD24 and CD44. Cells expressing SOX2 also showed down-regulated expression of the intestinal epithelial marker CDX2. We next evaluated CDX2 expression in our patient cohort. CDX2 down-regulation was more often found in right sided tumors of high grade and high stage. Furthermore, a decreased expression of CDX2 was closely linked to MSI, CIMP-high as well as BRAF mutated tumors. A decreased expression of CDX2 was also, in a stepwise manner, strongly correlated to a poor patient prognosis. When looking at SOX2 expression in relation to CDX2, we found that SOX2 expressing tumors more often displayed a down-regulated expression of CDX2. In addition, SOX2 expressing tumors with a down-regulated CDX2 expression had a worse patient prognosis compared to those with retained CDX2 expression. CONCLUSIONS: Our results indicate that SOX2 expression induces a cellular stem cell state in human CRC with a decreased expression of CDX2. Furthermore, a down-regulated expression of CDX2 results in a poor patient prognosis in CRC and at least part of the prognostic importance of SOX2 is mediated through CDX2 down-regulation.

Lundberg, I. V., et al. (2014). "SOX2 expression is regulated by BRAF and contributes to poor patient prognosis in colorectal cancer." PLoS One**9**(7): e101957.

Sporadic colorectal cancer (CRC) is a common malignancy and also one of the main causes of cancer deaths worldwide. Aberrant expression of the transcription factor SOX2 has recently been observed in several cancer types, but its role in CRC has not been fully elucidated. Here we studied the expression of SOX2 in 441 CRC patients by immunohistochemistry and related the expression to clinicopathological and molecular variables and patient prognosis. SOX2 was expressed in 11% of the tumors and was significantly associated to BRAFV600E mutation, but not to KRAS mutations (codon 12 and 13). SOX2 positivity was correlated to poor patient survival, especially in BRAFV600E mutated cases. In vitro studies showed that cells expressing the constitutively active BRAFV600E had increased SOX2 expression, a finding not found in cells expressing KRASG12V. Furthermore, blocking downstream BRAF signalling using a MEK-inhibitor resulted in a decreased expression of SOX2. Since SOX2 overexpression has been correlated to increased migration and invasion, we investigated the SOX2 expression in human CRC liver metastasis and found that a SOX2 positive primary CRC also had SOX2 expression in corresponding liver metastases. Finally we found that cells overexpressing SOX2 in vitro showed enhanced expression of FGFR1, which has been reported to correlate with liver metastasis in CRC. Our novel findings suggest that SOX2 expression is partly regulated by BRAF signalling, and an increased SOX2 expression may promote CRC metastasis and mediate a poor patient prognosis.

Luo, J., et al. (2018). "SOX2 inhibits cell proliferation and metastasis, promotes apoptotic by downregulating CCND1 and PARP in gastric cancer." Am J Transl Res**10**(2): 639-647.

Inconsistent results of Sex-determining region Y-box2 (SOX2) expression have been reported in gastric cancer (GC) before. Our recent studies showed that SOX2 was significantly downregulated in GC cells compared with GES-1 at both mRNA and protein level. Transfected with pcDNA3.1-SOX2 resulted in enforced expression of SOX2 at mRNA and protein levels compared with NC group in undifferentiated cell lines including HGC27 and BGC823. MTT assay showed that exogenous expressed SOX2 suppressed cell proliferation. FC analysis revealed that SOX2-overexpressing cells exhibited cell-cycle arrest and apoptosis. Transwell assay showed the anti-metastatic effect of SOX2 in GC cells. The subsequent results suggested CCND1 and PARP were downregulated in SOX2 overexpressed GC cells, and were responsible for the SOX2-induced anticancer effects. Thus, SOX2 proved to be an expected biomarker in GC diagnosis.

Maddison, P., et al. (2010). "Autoimmunity to SOX2, clinical phenotype and survival in patients with small-cell lung cancer."Lung Cancer**70**(3): 335-339.

OBJECTIVE: Autoantibodies to SOXB1 antigens are commonly found in patients with small-cell lung cancer (SCLC). It has not been established whether the presence of circulating SOX antibodies is associated with a specific paraneoplastic clinical phenotype, or if a tumour immune response to SOX antigens can affect prognosis in patients with SCLC in relation to other established prognostic factors. METHODS: Using recombinant SOX2 in an ELISA, we analysed sera in a prospective study from 212 unselected SCLC patients, which included 35 patients with neurological paraneoplastic disorders, or other well characterised onconeural antibodies. RESULTS: Overall, SOX2 antibodies were detected in 70 SCLC patients, with a sensitivity of 33% (95% CI 27-40%) and specificity of 97% (95% CI 94-99%) compared to controls matched for age, gender and smoking history.

No single clinical phenotype was seen in relation to the presence of SOX2 antibodies in isolation. Multivariate analysis showed that the presence of SOX2 antibodies in SCLC patients without evidence of neurological paraneoplastic disorders or onconeural antibodies did not have a significant effect on survival when known prognostic factors were accounted for. CONCLUSIONS: SOX2 antibodies are very specific markers for SCLC compared to matched non-tumour controls, but their presence does not seem to alter prognosis in this tumour type.

Maddison, P., et al. (2019). "The utility of anti-SOX2 antibodies for cancer prediction in patients with paraneoplastic neurological disorders."J Neuroimmunol**326**: 14-18.

Antibodies to SOXB1 proteins in patients with paraneoplastic disorders are associated with small-cell lung cancer (SCLC), particularly in Lambert-Eaton myasthenic syndrome (LEMS). We aimed to establish if SOX2 antibodies could be used to identify SCLC and other tumours found in a range of paraneoplastic disorders and controls. SOX2 antibodies were detectable in 61% of patients with LEMS-SCLC, and in other paraneoplastic disorders, such as opsoclonus-myoclonus and paraneoplastic cerebellar degeneration, only when there was an underlying SCLC. SOX2 antibodies are specific (>90%) markers for SCLC, but are rarely found in patients with other tumours, whether neurological symptoms are present or not.

Maehara, R., et al. (2018). "SOX2-silenced squamous cell carcinoma: a highly malignant form of esophageal cancer with SOX2 promoter hypermethylation." Mod Pathol**31**(1): 83-92.

This study originally aimed to investigate whether the overexpression of SOX2 is associated with the poor prognosis of patients with squamous cell carcinoma of the esophagus. However, we unexpectedly found that esophageal squamous cell carcinomas completely lacking SOX2 expression showed distinct pathologic features and highly aggressive clinical courses. The study cohort consisted of 113 consecutive patients with esophageal squamous cell carcinoma who underwent surgical resection without neoadjuvant therapy. Immunostaining on tissue microarrays and whole sections revealed that 8/113 (7%) cases were entirely negative for this transcriptional factor. SOX2-negative cancers were histologically less differentiated (P=0.002) and showed higher pT and pStages (P=0.003 and 0.007, respectively) than SOX2-positive cases. A remarkable finding was widespread lymphatic infiltration distant from the primary invasive focus, which was observed in 4 SOX2-negative cancers (50%), but none of the SOX2-positive cases. All separate dysplastic lesions observed in SOX2-negative cases were also SOX2-negative. The negative expression of SOX2 appeared to be an independent poor prognostic factor (OR=7.05, 95% CI=1.27-39.0). No mutations were identified in the coding or non-coding regions of SOX2. Fluorescent in situ hybridization did not show any copy-number variations in this gene. Since the SOX2 promoter contains an extensive CpG island, SOX2-negative cases underwent methylation-specific PCR, which disclosed promoter hypermethylation in all cases. In conclusion, SOX2-silenced squamous cell carcinomas of the esophagus appear to be a minor, but distinct form of malignancy characterized by extensive lymphatic invasion, a poor prognosis, and potential association with multiple SOX2-negative neoplastic lesions. The hypermethylation of the promoter region is seemingly a critical epigenetic event leading to SOX2 silencing.

Mamun, M. A., et al. (2018). "SOX2 in Cancer Stemness: Tumor Malignancy and Therapeutic Potentials." J Mol Cell Biol.

Cancer stem cells (CSCs), a minor subpopulation of tumor bulks with self-renewal and seeding capacity to generate new tumors, posit a significant challenge to develop effective and long-lasting anti-cancer therapies. The emergence of drug resistance appears upon failure of chemo-/radiation therapy to eradicate the CSCs, thereby leading to CSC-mediated clinical relapse. Accumulating evidence suggests that transcription factor SOX2, a master regulator of embryonic and induced pluripotent stem cells, drives cancer stemness, fuels tumor initiation, and contributes to tumor aggressiveness through major drug resistance mechanisms like epithelial-to-mesenchymal transition (EMT), ATP-binding cassette (ABC) drug transporters, anti-apoptotic and/or pro-survival signaling, lineage plasticity, and evasion of immune surveillance. Gaining a better insight and comprehensive interrogation into the mechanistic basis of SOX2-mediated generation of CSCs and treatment failure might therefore lead to new therapeutic targets involving CSC-specific anticancer strategies.

Matsika, A., et al. (2015). "Cancer stem cell markers in prostate cancer: an immunohistochemical study of ALDH1, SOX2 and EZH2." Pathology**47**(7): 622-628.

The aims of this study were to investigate the immunohistochemical expression and potential prognostic significance of putative cancer stems cell markers ALDH1, EZH2 and SOX2 in prostate cancer.A total of 142 consecutive radical prostatectomies submitted to one laboratory with a diagnosis of prostatic adenocarcinoma between 2008 and 2012 were retrieved and retrospectively studied.

Immunohistochemistry for the three markers was performed in each case and both univariate and multivariate analyses were undertaken to evaluate the correlation between the staining patterns and known histopathological prognostic features.ALDH1 showed a statistically significant association with tumour stage $p < 0.001$), extraprostatic extension ($p < 0.001$) and lymphovascular invasion ($p = 0.001$). EZH2 correlated with Gleason score ($p = 0.044$) and lymph node metastases ($p = 0.023$). SOX2 showed a statistically significant correlation with lymphovascular invasion only ($p = 0.018$) in both univariate and multivariate analyses.Cancer stem cell markers are variably expressed in prostate adenocarcinoma and immunohistochemical staining for ALDH1 and EZH2 may have a role in predicting tumour aggressiveness before treatment of prostate cancer.

Maurizi, G., et al. (2018). "Sox2 is required for tumor development and cancer cell proliferation in osteosarcoma." Oncogene**37**(33): 4626-4632.

The stem cell transcription factor Sox2 is highly expressed in many cancers where it is thought to mark cancer stem cells (CSCs). In osteosarcomas, the most common bone malignancy, high Sox2 expression marks and maintains a fraction of tumor-initiating cells that show all the properties of CSC. Knockdown of Sox2 expression abolishes tumorigenicity and suppresses the CSC phenotype. Here we show that, in a mouse model of osteosarcoma, osteoblast-specific Sox2 conditional knockout (CKO) causes a drastic reduction in the frequency and onset of tumors. The rare tumors detected in the Sox2 CKO animals were all Sox2 positive, indicating that they arose from cells that had escaped Sox2 deletion. Furthermore, Sox2 inactivation in cultured osteosarcoma cells by CRISPR/CAS technology leads to a loss of viability and proliferation of the entire cell population. Inactivation of the YAP gene, a major Hippo pathway effector which is a direct Sox2 target, causes similar results and YAP overexpression rescues cells from the lethality caused by Sox2 inactivation. These effects were osteosarcoma-specific, suggesting a mechanism of cell "addiction" to Sox2-initiated pathways. The requirement of Sox2 for osteosarcoma formation as well as for the survival of the tumor cells suggests that disruption of Sox2-initiated pathways could be an effective strategy for the treatment of osteosarcoma.

McCaughan, F., et al. (2010). "Progressive 3q amplification consistently targets SOX2 in preinvasive squamous lung cancer." Am J Respir Crit Care Med**182**(1): 83-91.

RATIONALE: Amplification of distal 3q is the most common genomic aberration in squamous lung cancer (SQC). SQC develops in a multistage progression from normal bronchial epithelium through dysplasia to invasive disease. Identifying the key driver events in the early pathogenesis of SQC will facilitate the search for predictive molecular biomarkers and the identification of novel molecular targets for chemoprevention and therapeutic strategies. For technical reasons, previous attempts to analyze 3q amplification in preinvasive lesions have focused on small numbers of predetermined candidate loci rather than an unbiased survey of copy-number variation. OBJECTIVES: To perform a detailed analysis of the 3q amplicon in bronchial dysplasia of different histological grades. METHODS: We use molecular copy-number counting (MCC) to analyze the structure of chromosome 3 in 19 preinvasive bronchial biopsy specimens from 15 patients and sequential biopsy specimens from 3 individuals. MEASUREMENTS AND MAIN RESULTS: We demonstrate that no low-grade lesions, but all high-grade lesions, have 3q amplification. None of seven low-grade lesions progressed clinically, whereas 8 of 10 patients with high-grade disease progressed to cancer. We identify a minimum commonly amplified region on chromosome 3 consisting of 17 genes, including 2 known oncogenes, SOX2 and PIK3CA. We confirm that both genes are amplified in all high-grade dysplastic lesions tested. We further demonstrate, in three individuals, that the clinical progression of high-grade preinvasive disease is associated with incremental amplification of SOX2, suggesting this promotes malignant progression. CONCLUSIONS: These findings demonstrate progressive 3q amplification in the evolution of preinvasive SQC and implicate SOX2 as a key target of this dynamic process.

Migita, T., et al. (2017). "Epithelial-mesenchymal transition promotes SOX2 and NANOG expression in bladder cancer." Lab Invest.

Bladder cancer is the most common malignant tumor of the urothelium and is classified into non-muscle-invasive bladder cancer (NMIBC) and muscle-invasive bladder cancer (MIBC). Stemness markers such as SOX2 and NANOG are frequently overexpressed in various aggressive cancers, including MIBC; epithelial-mesenchymal transition (EMT) has been proposed as a potential trigger of stemness in cancers. To determine whether cancer stemness is acquired via EMT in bladder cancer, we studied the effect of EMT on the expression of SOX2 and NANOG in bladder cancer cell lines. We also analyzed their expression in clinical tissue samples. Our results revealed that a potent EMT inducer (transforming growth factor beta1) reduced the expression of the epithelial marker E-cadherin and increased expression of both SOX2 and NANOG in epithelial-type bladder cancer cells. As for clinical

bladder cancer samples, in NMIBC, E-cadherin expression was slightly diminished, and the expression of both SOX2 and NANOG was negligible. In contrast, in MIBC, E-cadherin expression was highly and heterogeneously diminished, while the expression of both SOX2 and NANOG was increased. We also noticed that either E-cadherin or SOX2 (or NANOG) was expressed (ie, in a manner exclusive of each other). In addition, the concentration of E-cadherin showed a significant negative correlation with tumor grade and stage, while expression of SOX2 and NANOG positively correlated with those clinicopathological parameters. These findings suggest that EMT promotes stemness of bladder cancer cells, contributing to tumor aggressiveness. This EMT-cancer stemness axis may also play an important role in the pathogenesis of NMIBC and MIBC.Laboratory Investigation advance online publication, 27 February 2017; doi:10.1038/labinvest.2017.17.

Miller, T. J., et al. (2017). "The prognostic value of cancer stem-like cell markers SOX2 and CD133 in stage III colon cancer is modified by expression of the immune-related markers FoxP3, PD-L1 and CD3." Pathology**49**(7): 721-730.

Cancer stem-like cells are highly tumourigenic cells that can repopulate entire tumours after apparent successful treatment. Recent evidence suggests they interact with other cells in the tumour microenvironment, including immune cell subsets, to enhance their survival. The aim of this study was to determine whether the expression of immune cell markers in primary colon cancer impacts the prognostic significance of cancer stem-like cell marker expression. Immunohistochemistry was used to assess the expression of putative stem cell markers (ALDH1, CD44v6, CD133, Lgr5, SOX2) and immune cell related markers (CD3, CD8, FoxP3, PD-L1) in 104 patients with stage III colon cancer. Associations of marker expression with overall and cancer-specific survival were determined using Kaplan-Meier analysis. High SOX2 expression in the central tumour area was found to be an independent factor for poor cancer-specific survival [hazard ratio (HR) 6.19; 95% confidence interval (CI) 2.24-17.14; p=0.001]. When immune-related factors were taken into account, patients categorised as SOX2(low)/FoxP3(high) had good outcome (HR 0.164; 95%CI 0.066-0.406; p<0.0001) whereas patients categorised as SOX2(high)/PD-L1(low) had poor outcome (HR 8.992; 95%CI 3.397-23.803; p<0.0001). The prognostic value of the SOX2 cancer stem-like cell marker in colon cancer is modified by expression of immune-cell related factors FoxP3 and PD-L1.

Mollaoglu, G., et al. (2018). "The Lineage-Defining Transcription Factors SOX2 and NKX2-1 Determine Lung Cancer Cell Fate and Shape the Tumor Immune Microenvironment." Immunity**49**(4): 764-779 e769.

The major types of non-small-cell lung cancer (NSCLC)-squamous cell carcinoma and adenocarcinoma-have distinct immune microenvironments. We developed a genetic model of squamous NSCLC on the basis of overexpression of the transcription factor Sox2, which specifies lung basal cell fate, and loss of the tumor suppressor Lkb1 (SL mice). SL tumors recapitulated gene-expression and immune-infiltrate features of human squamous NSCLC; such features included enrichment of tumor-associated neutrophils (TANs) and decreased expression of NKX2-1, a transcriptional regulator that specifies alveolar cell fate. In Kras-driven adenocarcinomas, mis-expression of Sox2 or loss of Nkx2-1 led to TAN recruitment. TAN recruitment involved SOX2-mediated production of the chemokine CXCL5. Deletion of Nkx2-1 in SL mice (SNL) revealed that NKX2-1 suppresses SOX2-driven squamous tumorigenesis by repressing adeno-to-squamous transdifferentiation. Depletion of TANs in SNL mice reduced squamous tumors, suggesting that TANs foster squamous cell fate. Thus, lineage-defining transcription factors determine the tumor immune microenvironment, which in turn might impact the nature of the tumor.

Mou, W., et al. (2015). "Expression of Sox2 in breast cancer cells promotes the recruitment of M2 macrophages to tumor microenvironment." Cancer Lett**358**(2): 115-123.

Transcriptional factor Sox2 promotes tumor metastasis; however its regulatory effect on tumor-associated macrophages (TAMs, M2 phenotype) has not been defined. This study disclosed concomitant expression of TAMs marker-CD163 with SOX2 in human breast cancer and showed that Sox2 in breast cancer cells promotes recruitment of TAMs with altered expression of multiple chemokines, including MIP-1alpha, ICAM-1 etc. and activation of Stat3 and NF-kappaB signalings. In addition, TAMs rescued the compromised lung metastasis induced by Sox2 silencing in breast cancer cells. Together, this study documented that Sox2 plays an important role in recruiting TAMs and promotes tumor metastasis in a TAMs dependent manner.

Mu, P., et al. (2017). "SOX2 promotes lineage plasticity and antiandrogen resistance in TP53- and RB1-deficient prostate cancer." Science**355**(6320): 84-88.

Some cancers evade targeted therapies through a mechanism known as lineage plasticity, whereby tumor cells acquire phenotypic characteristics of a cell lineage whose survival no longer depends on the drug target. We use in vitro and in vivo human prostate cancer models to show that these tumors can develop resistance to the antiandrogen drug enzalutamide by a phenotypic shift from androgen receptor (AR)-dependent luminal epithelial cells to AR-independent basal-like cells. This lineage plasticity is enabled by the loss of TP53 and RB1 function, is mediated by increased expression of the reprogramming transcription factor SOX2, and can be reversed by restoring TP53 and RB1 function or by inhibiting SOX2 expression. Thus, mutations in tumor suppressor genes can create a state of increased cellular plasticity that, when challenged with antiandrogen therapy, promotes resistance through lineage switching.

Mukherjee, P., et al. (2017). "Modulation of SOX2 expression delineates an end-point for paclitaxel-effectiveness in breast cancer stem cells." Sci Rep**7**(1): 9170.

Tumor relapse in triple negative breast cancer patients has been implicated to chemoresistant cancer stem cells (CSCs), which under favorable conditions culminate in tumor re-formation and metastasis. Hence, eradication of CSCs during systemic chemotherapy is imperative. CSCs were sorted using immuno-phenotyping and aldefluor assay. Gene expression profiling of normal breast stem cells and breast CSCs from chemo-treated patients were carried out. Silencing SOX2 was achieved by siRNA method. Mammosphere culture and wound healing assays were carried out to assess efficacy of CSCs. Microarray analysis revealed elevated expression of SOX2, ABCG2 and TWIST1, unraveling an intertwined pluripotency-chemoresistance-EMT axis. Although paclitaxel treatment led to temporary arrest of cell migration, invasiveness resumed after drug removal. The 'twist in the tale' was a consistently elevated expression of TWIST1, substantiating that TWIST1 can also promote stemness and chemoresistance in tumors; hence, its eradication was imperative. Silencing SOX2 increased chemo-sensitivity and diminished sphere formation, and led to TWIST1 down regulation. This study eventually established that SOX2 silencing of CSCs along with paclitaxel treatment reduced SOX2-ABCG2-TWIST1 expression, disrupted sphere forming capacity and also reduced invasiveness by retaining epithelial-like properties of the cells, thereby suggesting a more comprehensive therapy for TNBC patients in future.

Mukhopadhyay, A., et al. (2014). "Sox2 cooperates with Lkb1 loss in a mouse model of squamous cell lung cancer." Cell Rep**8**(1): 40-49.

Squamous cell carcinoma (SCC) of the lung is the second most common subtype of lung cancer. With limited treatment options, the 5-year survival rate of SCC is only 15%. Although genomic alterations in SCC have been characterized, identifying the alterations that drive SCC is critical for improving treatment strategies. Mouse models of SCC are currently limited. Using lentiviral delivery of Sox2 specifically to the mouse lung, we tested the ability of Sox2 to promote tumorigenesis in multiple tumor suppressor backgrounds. Expression of Sox2, frequently amplified in human SCC, specifically cooperates with loss of Lkb1 to promote squamous lung tumors. Mouse tumors exhibit characteristic histopathology and biomarker expression similar to human SCC. They also mimic human SCCs by activation of therapeutically relevant pathways including STAT and mTOR. This model may be utilized to test the contribution of additional driver alterations in SCC, as well as for preclinical drug discovery.

Mukhopadhyay, A. and T. G. Oliver (2015). "Mighty mouse breakthroughs: a Sox2-driven model for squamous cell lung cancer." Mol Cell Oncol**2**(2): e969651.

Squamous lung cancer is a subtype of non-small cell lung cancer with a poor overall prognosis. We have recently generated a mouse model of squamous lung carcinoma by overexpressing Sex-determining region Y-box 2 (Sox2) and deleting liver kinase B1 (Lkb1) using a lentiviral approach. This model recapitulates the human disease in terms of histopathology, biomarker expression, and signaling pathway activation, making it an excellent model for preclinical studies.

Neumann, J., et al. (2011). "SOX2 expression correlates with lymph-node metastases and distant spread in right-sided colon cancer." BMC Cancer**11**: 518.

BACKGROUND: The transcription factor SOX2, which is involved in the induction of pluripotent stem cells and contributes to colorectal carcinogenesis, is associated with a poor prognosis in colon cancer (CC). Furthermore, SOX2 is a repressor of the transcriptional activity of beta-catenin in vitro. Since the majority of CC develop via an activation of the Wnt/beta-catenin signalling pathway, indicated by nuclear expression of beta-catenin, we wanted to investigate the expression patterns of SOX2 and beta-catenin and correlate them with the occurrence of lymph node and distant metastases as indicators of

malignant progression. METHODS: The expression of SOX2 and beta-catenin was investigated in a case control study utilizing a matched pair collection (N = 114) of right-sided CCs with either corresponding distant metastases (N = 57) or without distant spread (N = 57) by applying immunohistochemistry. RESULTS: Elevated protein expression of SOX2 significantly correlated with the presence of lymph node- (p = 0.006) and distant metastases (p = 0.022). Nuclear beta-catenin expression correlated significantly only with distant metastases (p = 0.001). Less than 10% of cases showed a coexpression of high levels of beta-catenin and SOX2. The positivity for both markers was also associated with a very high risk for lymph-node metastases (p = 0.007) and distant spread (p = 0.028). CONCLUSION: We demonstrated that increased expression of either SOX2 or nuclear beta-catenin are associated with distant metastases in right-sided CC. Additionally, SOX2 is also associated with lymph-node metastases. These data underline the importance of stemness-associated markers for the identification of CC with high risk for distant spread.

Ooki, A., et al. (2018). "Epigenetically regulated PAX6 drives cancer cells toward a stem-like state via GLI-SOX2 signaling axis in lung adenocarcinoma." Oncogene**37**(45): 5967-5981.

It remains unclear whether PAX6 acts as a crucial transcription factor for lung cancer stem cell (CSC) traits. We demonstrate that PAX6 acts as an oncogene responsible for induction of cancer stemness properties in lung adenocarcinoma (LUAD). Mechanistically, PAX6 promotes GLI transcription, resulting in SOX2 upregulation directly by the binding of GLI to the proximal promoter region of the SOX2 gene. The overexpressed SOX2 enhances the expression of key pluripotent factors (OCT4 and NANOG) and suppresses differentiation lineage factors (HOPX and NKX2-1), driving cancer cells toward a stem-like state. In contrast, in the differentiated non-CSCs, PAX6 is transcriptionally silenced by its promoter methylation. In human lung cancer tissues, the positive linear correlations of PAX6 expression with GLI and SOX2 expression and its negative correlations with HOPX and NKX2-1 expression were observed. Therapeutically, the blockade of the PAX6-GLI-SOX2 signaling axis elicits a long-lasting therapeutic efficacy by limiting CSC expansion following chemotherapy. Furthermore, a methylation panel including the PAX6 gene yielded a sensitivity of 79.1% and specificity of 83.3% for cancer detection using serum DNA from stage IA LUAD. Our findings provide a rationale for targeting the PAX6-GLI-SOX2 signaling axis with chemotherapy as an effective therapeutic strategy and support the clinical utility of PAX6 gene promoter methylation as a biomarker for early lung cancer detection.

Ozen, M., et al. (2015). "Overexpression of miR-145-5p inhibits proliferation of prostate cancer cells and reduces SOX2 expression." Cancer Invest**33**(6): 251-258.

We aimed to perform functional analysis of miR-145-5p in prostate cancer (PCa) cells and to identify targets of miR-145-5p for understanding its role in PCa pathogenesis. PC3, DU145, LNCaP PCa, and PNT1a nontumorigenic prostate cell lines were utilized for functional analysis of miR-145-5p. Its overexpression caused inhibition of proliferation through apoptosis and reduced migration in PCa cells. SOX2 expression was significantly decreased in both mRNA and protein level in miR-145-5p-overexpressed PCa cells. We proposed that miR-145-5p, being an important regulator of SOX2, carries a crucial role in PCa tumorigenesis.

Park, G. B. and D. Kim (2017). "TLR5/7-mediated PI3K activation triggers epithelial-mesenchymal transition of ovarian cancer cells through WAVE3-dependent mesothelin or OCT4/SOX2 expression." Oncol Rep**38**(5): 3167-3176.

Toll-like receptor (TLR)-mediated signaling induces cell migration or invasion in several tumors and various stages of cancer. Interactions of mesothelin, a 40-kDa cell surface glycoprotein, with cancer antigen 125 (CA125) is associated with drug resistance, metastasis, and poor clinical outcome of ovarian cancer patients. In this study, we examined the role of TLR5 and TLR7 in the metastasis of ovarian cancer through the induction of mesothelin/CA125 expression and investigated its underlying mechanism. TLR5 agonist (flagellin) and TLR7 agonist (imiquimod) upregulated mesenchymal phenotypes and produced epithelial-mesenchymal transition (EMT)-related cytokines in the SKOV3 cells; however, TLR7 expressing CaOV3 cells had no response to the specific ligand, imiquimod, for enhancing its EMT processes. Stimulation of the SKOV3 cells with flagellin or imiquimod activated Wiskott-Aldrich syndrome protein verprolin-homologous 3 (WAVE3) and mesothelin/CA125, whereas it suppressed the expression of TAp63. Moreover, knockdown of TLR5 or TLR7 in SKOV3 cells profoundly impaired the TLR5- or TLR7-intiated downstream signaling pathway. Loss of WAVE3 in SKOV3 cells led to the inhibition of invasion, suppression of mesenchymal characteristics, prevention of OCT4/SOX2 secretion, and attenuation of mesothelin/CA125 expression after stimulation with flagellin or imiquimod. Although the disruption of

mesothelin decreased the migratory activity of the TLR5/7-activated SKOV3 cells, knockdown of mesothelin failed to reduce the expression of mesenchymal markers, OCT4, and SOX2. In addition, targeting OCT4 or SOX2 with siRNA had no effect on the expression of mesothelin and the suppression of transcriptionally active p63 (TAp63) in the TLR5/7-stimulated SKOV3 cells. Our results suggest that TLR5/7-mediated WAVE3 activation not only controls the mesothelin-related EMT processes but also modulates OCT4/SOX2-mediated mesenchymal marker expression. Taken together, both TLR5 and TLR7 expression are critical for the TLR5/7-induced metastasis of ovarian cancer and the inhibition of WAVE3 might be a new therapeutic target to control ovarian cancer metastasis.

Phi, L. T. H., et al. (2018)."The anti-metastatic effect of ginsenoside Rb2 in colorectal cancer in an EGFR/SOX2-dependent manner."Cancer Med**7**(11): 5621-5631.

Ginsenoside Rb2, a saponin from Panax ginseng, has been shown to have many functions. However, the effect of ginsenoside Rb2 on the metastasis of colorectal cancer (CRC) remains unknown. CRC cell lines HT29 and SW620 were used to determine the effects of ginsenoside Rb2 on the colony-forming, migration, invasion, and wound-healing abilities of CRC cells in vitro. Further, ginsenoside Rb2 was given intraperitoneally at 5 mg/kg of mouse body weight to check its effect on the metastasis of CRC cells in vivo. Ginsenoside Rb2 decreased colony-forming ability, migration, invasion, and wound healing of CRC cells in vitro, although it did not affect cell proliferation. As a possible mechanism, we found that ginsenoside Rb2 down-regulated the expression of stemness and Epithelial-mesenchymal transition (EMT)-related genes via the EGFR/SOX2 signaling axis; these were partially rescued by either exogenous EGF treatment or ectopic expression of SOX2. More importantly, ginsenoside Rb2 significantly reduced the number of metastatic nodules in the livers, lungs, and kidneys in a mouse model of metastasis. These results suggest that ginsenoside Rb2 could be used to treat the metastasis of CRC therapeutically or as a supplement.

Picon-Ruiz, M., et al. (2016)."Interactions between Adipocytes and Breast Cancer Cells Stimulate Cytokine Production and Drive Src/Sox2/miR-302b-Mediated Malignant Progression."Cancer Res**76**(2): 491-504.

Consequences of the obesity epidemic on cancer morbidity and mortality are not fully appreciated. Obesity is a risk factor for many cancers, but the mechanisms by which it contributes to cancer development and patient outcome have yet to be fully elucidated. Here, we examined the effects of coculturing human-derived adipocytes with established and primary breast cancer cells on tumorigenic potential. We found that the interaction between adipocytes and cancer cells increased the secretion of proinflammatory cytokines. Prolonged culture of cancer cells with adipocytes or cytokines increased the proportion of mammosphere-forming cells and of cells expressing stem-like markers in vitro. Furthermore, contact with immature adipocytes increased the abundance of cancer cells with tumor-forming and metastatic potential in vivo. Mechanistic investigations demonstrated that cancer cells cultured with immature adipocytes or cytokines activated Src, thus promoting Sox2, c-Myc, and Nanog upregulation. Moreover, Sox2-dependent induction of miR-302b further stimulated cMYC and SOX2 expression and potentiated the cytokine-induced cancer stem cell-like properties. Finally, we found that Src inhibitors decreased cytokine production after coculture, indicating that Src is not only activated by adipocyte or cytokine exposures, but is also required to sustain cytokine induction. These data support a model in which cancer cell invasion into local fat would establish feed-forward loops to activate Src, maintain proinflammatory cytokine production, and increase tumor-initiating cell abundance and metastatic progression. Collectively, our findings reveal new insights underlying increased breast cancer mortality in obese individuals and provide a novel preclinical rationale to test the efficacy of Src inhibitors for breast cancer treatment.

Piva, M., et al. (2014). "Sox2 promotes tamoxifen resistance in breast cancer cells." EMBO Mol Med**6**(1): 66-79.

Development of resistance to therapy continues to be a serious clinical problem in breast cancer management. Cancer stem/progenitor cells have been shown to play roles in resistance to chemo- and radiotherapy. Here, we examined their role in the development of resistance to the oestrogen receptor antagonist tamoxifen. Tamoxifen-resistant cells were enriched for stem/progenitors and expressed high levels of the stem cell marker Sox2. Silencing of the SOX2 gene reduced the size of the stem/progenitor cell population and restored sensitivity to tamoxifen. Conversely, ectopic expression of Sox2 reduced tamoxifen sensitivity in vitro and in vivo. Gene expression profiling revealed activation of the Wnt signalling pathway in Sox2-expressing cells, and inhibition of Wnt signalling sensitized resistant cells to tamoxifen. Examination of patient tumours indicated that Sox2 levels are higher in patients after endocrine therapy failure, and also in the primary tumours of these patients, compared to those of responders.

Together, these results suggest that development of tamoxifen resistance is driven by Sox2-dependent activation of Wnt signalling in cancer stem/progenitor cells.

Qin, J., et al. (2015). "DC120, a novel AKT inhibitor, preferentially suppresses nasopharyngeal carcinoma cancer stem-like cells by downregulating Sox2." Oncotarget**6**(9): 6944-6958.

Side population (SP) contains cancer stem-like cells (CSLCs). In this study, we characterized SP cells from nasopharyngeal carcinoma (NPC) cell lines and found that SP cells had a higher self-renewal ability in vitro and greater tumorigenicity in vivo. The AKT pathway was activated in NPC SP cells. DC120, a 2-pyrimidyl-5-amidothiazole inhibitor of the ATP binding site of AKT, inhibited phosphorylation of FKHRL1 and GSK-3beta. DC120 inhibited SP fraction, the sphere-forming ability in vitro and growth of primary xenografts as well as secondary xenografts' tumor recurrence. This inhibition was accompanied by reduced expression of stem-related gene Sox2 due to induction of p27 and miR-30a. A combination of DC120 and CDDP more effectively inhibited NPC cells compared with monotherapy in vitro and in vivo. Clinical evaluation of DC120 is warranted.

Riggi, N., et al. (2010). "EWS-FLI-1 modulates miRNA145 and SOX2 expression to initiate mesenchymal stem cell reprogramming toward Ewing sarcoma cancer stem cells." Genes Dev**24**(9): 916-932.

Cancer stem cells (CSCs) display plasticity and self-renewal properties reminiscent of normal tissue stem cells, but the events responsible for their emergence remain obscure. We recently identified CSCs in Ewing sarcoma family tumors (ESFTs) and showed that they retain mesenchymal stem cell (MSC) plasticity. In the present study, we addressed the mechanisms that underlie ESFT CSC development. We show that the EWS-FLI-1 fusion gene, associated with 85%-90% of ESFTs and believed to initiate their pathogenesis, induces expression of the embryonic stem cell (ESC) genes OCT4, SOX2, and NANOG in human pediatric MSCs (hpMSCs) but not in their adult counterparts. Moreover, under appropriate culture conditions, hpMSCs expressing EWS-FLI-1 generate a cell subpopulation displaying ESFT CSC features in vitro. We further demonstrate that induction of the ESFT CSC phenotype is the result of the combined effect of EWS-FLI-1 on its target gene expression and repression of microRNA-145 (miRNA145) promoter activity. Finally, we provide evidence that EWS-FLI-1 and miRNA-145 function in a mutually repressive feedback loop and identify their common target gene, SOX2, in addition to miRNA145 itself, as key players in ESFT cell differentiation and tumorigenicity. Our observations provide insight for the first time into the mechanisms whereby a single oncogene can reprogram primary cells to display a CSC phenotype.

Rodriguez-Pinilla, S. M., et al. (2007). "Sox2: a possible driver of the basal-like phenotype in sporadic breast cancer." Mod Pathol**20**(4): 474-481.

Tumours arising in BRCA1 mutation carriers and sporadic basal-like breast carcinomas have similar phenotypic, immunohistochemical and clinical characteristics. SOX2 is an embryonic transcription factor located at chromosome 3q, a region frequently gained in sporadic basal-like and BRCA1 germline mutated tumours. The aim of the study was to establish whether sox2 expression was related to basal-like sporadic breast tumours. Two hundred and twenty-six sporadic node-negative invasive breast carcinomas were immunohistochemically analysed for oestrogen receptor (ER), progesterone receptor (PR), CK5/6, EGFR, vimentin, HER2, ki67, p53 and sox2 using tissue microarrays. Tumours were considered to have basal-like phenotype if they were ER/HER2-negative and CK5/6 and/or EGFR-positive. Thirty cases of this series (13.7%) displayed a basal-like phenotype. Sox2 expression was observed in 16.7% of cases and was significantly more frequently expressed in basal-like breast carcinomas (43.3% in basal-like, 10.6% in luminal and 13.3% in HER2+ tumours, $P<0.001$). Moreover, Sox2 showed a statistically significant inverse association with ER and PR ($P=0.001$ and 0.017, respectively) and direct association with CK5/6, EGFR and vimentin ($P=0.022$, 0.005 and <0.001, respectively). Sox2 is preferentially expressed in tumours with basal-like phenotype and may play a role in defining their less differentiated/'stem cell' phenotypic characteristics.

Rothenberg, S. M., et al. (2015). "Inhibition of mutant EGFR in lung cancer cells triggers SOX2-FOXO6-dependent survival pathways." Elife**4**.

Treatment of EGFR-mutant lung cancer with erlotinib results in dramatic tumor regression but it is invariably followed by drug resistance. In characterizing early transcriptional changes following drug treatment of mutant EGFR-addicted cells, we identified the stem cell transcriptional regulator SOX2 as being rapidly and specifically induced, both in vitro and in vivo. Suppression of SOX2 sensitizes cells to erlotinib-mediated apoptosis, ultimately decreasing the emergence of acquired resistance, whereas its ectopic expression reduces drug-induced cell death. We show that erlotinib relieves EGFR-dependent suppression of FOXO6, leading to its induction of SOX2, which in turn represses the pro-apoptotic BH3-only genes BIM and BMF. Together, these observations point to a physiological feedback mechanism that attenuates

oncogene addiction-mediated cell death associated with the withdrawal of growth factor signaling and may therefore contribute to the development of resistance.

Ruan, J., et al. (2013). "Predictive value of Sox2 expression in transurethral resection specimens in patients with T1 bladder cancer."Med Oncol**30**(1): 445.

Sox2 is thought to be an important regulator of self-renewal in embryonic stem cell. According to the cancer stem cell (CSC) theory, the overexpression of Sox2 is potentially involved in carcinogenesis and could affect tumor recurrence and metastasis. Previous study proved Sox2 might be prognostic marker for multiple human malignancies. The purpose of this study was to investigate the clinicopathological significance of Sox2 expression in human non-muscle-invasive bladder cancer. We examined Sox2 expression in 32 paired non-muscle-invasive bladder cancer tissues and adjacent non-cancerous tissues by quantitative real-time RT-PCR (qrtRT-PCR). In addition, we analyzed Sox2 and Ki-67 expression in 126 non-muscle-invasive bladder cancer samples and bladder cancer cell line T24 by immunohistochemistry and immunofluorescence assays. The recurrence-free survival was determined by Kaplan-Meier method and log-rank test. Cox regression was adopted for univariate and multivariate analyses of prognostic factors. The expression of Sox2 was significantly increased in non-muscle-invasive bladder cancer tissues. Sox2 expression was significantly correlated with that of Ki-67 ($P < 0.001$). The expression of Sox2 was significantly associated with tumor size ($P = 0.006$), tumor number ($P = 0.037$), and tumor grade ($P < 0.001$). Patients with high Sox2 expression had significantly poorer recurrence-free survival ($P = 0.0002$) when compared with patients with the low expression of Sox2. On multivariate analysis, Sox2 expression and tumor grade were found to be independent prognostic factors for recurrence-free survival ($P < 0.05$). Our data suggested for the first time that the high expression of Sox2 may contribute to the development of non-muscle-invasive bladder cancer and serve as a novel prognostic marker in patients with T1 bladder cancer.

Rudin, C. M., et al. (2012). "Comprehensive genomic analysis identifies SOX2 as a frequently amplified gene in small-cell lung cancer." Nat Genet**44**(10): 1111-1116.

Small-cell lung cancer (SCLC) is an exceptionally aggressive disease with poor prognosis. Here, we obtained exome, transcriptome and copy-number alteration data from approximately 53 samples consisting of 36 primary human SCLC and normal tissue pairs and 17 matched SCLC and lymphoblastoid cell lines. We also obtained data for 4 primary tumors and 23 SCLC cell lines. We identified 22 significantly mutated genes in SCLC, including genes encoding kinases, G protein-coupled receptors and chromatin-modifying proteins. We found that several members of the SOX family of genes were mutated in SCLC. We also found SOX2 amplification in approximately 27% of the samples. Suppression of SOX2 using shRNAs blocked proliferation of SOX2-amplified SCLC lines. RNA sequencing identified multiple fusion transcripts and a recurrent RLF-MYCL1 fusion. Silencing of MYCL1 in SCLC cell lines that had the RLF-MYCL1 fusion decreased cell proliferation. These data provide an in-depth view of the spectrum of genomic alterations in SCLC and identify several potential targets for therapeutic intervention.

Russo, M. V., et al. (2016). "SOX2 boosts major tumor progression genes in prostate cancer and is a functional biomarker of lymph node metastasis." Oncotarget**7**(11): 12372-12385.

Critical issues in prostate cancer (PC) are a. identification of molecular drivers of the highly aggressive neuroendocrine differentiation (NED) in adenocarcinoma, and b. early assessment of disease progression. The SRY (sex determining region Y)-box 2 gene, SOX2, is an essential embryonic stem cell gene involved in prostate tumorigenesis. Here we assessed its implications in NED and progression of PC and its diagnostic and prognostic value. Laser microdissection, qRT-PCR, quantitative Methylation-Specific PCR and immunohistochemistry were used to analyze SOX2 gene expression and regulation in 206 PC samples. Results were examined according to the patient's clinical pathological profile and follow-ups. Functional studies were performed using PC cells transfected to overexpress or silence SOX2. SOX2 was consistently downregulated in PC, except in cell clusters lying within lymph node (LN)-positive PC. Multivariate analysis revealed that SOX2 mRNA expression in the primary tumor was significantly associated with LN metastasis. When SOX2 mRNA levels were >/=1.00, relative to (XpressRef) Universal Total RNA, adjusted Odds Ratio was 24.4 (95% CI: 7.54-79.0), sensitivity 0.81 (95% CI: 0.61-0.93) and specificity 0.87 (95% CI: 0.81-0.91). Patients experiencing biochemical recurrence had high median levels of SOX2 mRNA. In both PC and LN metastasis, SOX2 and NED marker, Chromogranin-A, were primarily co-expressed. In PC cells, NED genes were upregulated by SOX2 overexpression and downregulated by its silencing, which also abolished SNAI2/Slug dependent NED. Moreover, SOX2 upregulated neural CAMs, neurotrophins/neurotrophin receptors, pluripotency and epithelial-mesenchymal

transition transcription factors, growth, angiogenic and lymphangiogenic factors, and promoted PC cell invasiveness and motility. This study discloses novel SOX2 target genes driving NED and spread of PC and proposes SOX2 as a functional biomarker of LN metastasization for PC.

Rybak, A. P. and D. Tang (2013). "SOX2 plays a critical role in EGFR-mediated self-renewal of human prostate cancer stem-like cells." Cell Signal**25**(12): 2734-2742.

SOX2 is an essential transcription factor for stem cells and plays a role in tumorigenesis, however its role in prostate cancer stem cells (PCSCs) remains unclear. We report here a significant upregulation of SOX2 at both mRNA and protein levels in DU145 PCSCs propagated as suspension spheres in vitro. The expression of SOX2 in DU145 PCSCs is positively regulated by epidermal growth factor receptor (EGFR) signaling. Activation of EGFR signaling, following the addition of epidermal growth factor (EGF) or ectopic expression of a constitutively-active EGFR mutant (EGFRvIII), increased SOX2 expression and the self-renewal of DU145 PCSCs. Conversely, a small molecule EGFR inhibitor (AG1478) blocked EGFR activation, reduced SOX2 expression and inhibited PCSC self-renewal activity, implicating SOX2 in mediating EGFR-dependent self-renewal of PCSCs. In line with this notion, ectopic SOX2 expression enhanced EGF-induced self-renewal of DU145 PCSCs, while SOX2 knockdown reduced PCSC self-renewal with EGF treatment no longer capable of enhancing their propagation. Furthermore, SOX2 knockdown reduced the capacity of DU145 PCSCs to grow under anchorage-independent conditions. Finally, DU145 PCSCs generated xenograft tumors more aggressively with elevated levels of SOX2 expression compared to xenograft tumors derived from non-PCSCs. Collectively, we provide evidence that SOX2 plays a critical role in EGFR-mediated self-renewal of DU145 PCSCs.

Saigusa, S., et al. (2009)."Correlation of CD133, OCT4, and SOX2 in rectal cancer and their association with distant recurrence after chemoradiotherapy."Ann Surg Oncol**16**(12): 3488-3498.

BACKGROUND: Cancer stem cells are associated with metastatic potential, treatment resistance, and poor patient prognosis. Distant recurrence remains the major cause of mortality in rectal cancer patients with preoperative chemoradiotherapy (CRT). We investigated the role of three stem cell markers (CD133, OCT4, and SOX2) in rectal cancer and evaluated the association between these gene levels and clinical outcome in rectal cancer patients with preoperative CRT. METHODS: Thirty-three patients with rectal cancer underwent preoperative CRT. Total RNAs of rectal cancer cells before and after CRT were isolated. Residual cancer cells after CRT were obtained from formalin-fixed paraffin-embedded (FFPE) specimens using microdissection. The expression levels of three stem cell genes were measured using real-time reverse-transcription polymerase chain reaction (RT-PCR). The association between these gene levels and radiation was evaluated using colon cancer cell lines. Immunohistochemical staining of these markers after CRT was also investigated. RESULTS: There were significant positive correlations among the three genes after CRT. Patients who developed distant recurrence had higher levels of the three genes compared with those without recurrence in residual cancer after CRT. These elevated gene levels were significantly associated with poor disease-free survival. The radiation caused upregulation of these gene levels in LoVo and SW480 in vitro. Immunohistochemically, CD133 staining was observed in not only luminal surface but also cytoplasm. CONCLUSIONS: Expression of CD133, OCT4, and SOX2 may predict distant recurrence and poor prognosis of rectal cancer patients treated with preoperative CRT. Correlations among these genes may be associated with tumor regrowth and metastatic relapse after CRT.

Samulin Erdem, J., et al. (2016). "Mutations in TP53 increase the risk of SOX2 copy number alterations and silencing of TP53 reduces SOX2 expression in non-small cell lung cancer." BMC Cancer**16**: 28.

BACKGROUND: Amplifications of the transcription factor, SRY (sex determining region Y)-box 2 (SOX2), are common in non-small cell lung cancer (NSCLC). SOX2 signaling is important in maintaining the stem cell-like phenotype of cancer cells and contributes to the pathogenesis of lung cancer. TP53 is known to inhibit gene amplifications and to repress many stem cell-associated genes following DNA damage. The aim of this study was to investigate if TP53 mutational status affected SOX2 copy number variation and gene expression in early-stage NSCLC patients; moreover, to assess if TP53 regulates SOX2 expression in human lung cancer cells. METHODS: 258 early-stage lung cancer patients were included in the study. Exons 4-9 in the TP53 gene were sequenced for mutations in tumor tissues. SOX2 copy number as well as TP53 and SOX2 gene expression were analyzed in tumor and in adjacent non-tumorous tissues by qPCR. TP53 and SOX2 were silenced using gene-specific siRNAs in human lung adenocarcinoma A427 cells, and the expression of TP53, SOX2 and subset of selected miRNAs was analyzed by qPCR. The odds ratios (ORs) for

associations between copy number variation and lung cancer were estimated by conditional logistic regression, and the correlation between gene status and clinicopathological characteristics was assessed by Chi-square or Fisher's exact test. Gene expression data was analyzed using nonparametric Mann-Whitney test. RESULTS: TP53 mutations were associated with an increased risk of acquiring a SOX2 copy number alteration (OR = 2.08, 95% CI: 1.14-3.79, p = 0.017), which was more frequently occurring in tumor tissues (34%) than in adjacent non-tumorous tissues (3%). Moreover, SOX2 and TP53 expression levels were strongly correlated in tumor tissues. In vitro studies showed that a reduction in TP53 was associated with decreased SOX2 expression in A427 cells. Furthermore, TP53 knockdown reduced the miRNA hsa-miR-145, which has previously been shown to regulate SOX2 expression. CONCLUSIONS: TP53 signaling may be important in the regulation of SOX2 copy number and expression in NSCLC tumors, and the miRNA hsa-miR-145-5p may be one potential driver. This prompts for further studies on the mechanisms behind the TP53-induced regulation of SOX2 expression and the possible importance of hsa-miR-145 in lung cancer.

Schaal, C. M., et al. (2018)."Regulation of Sox2 and stemness by nicotine and electronic-cigarettes in non-small cell lung cancer."Mol Cancer**17**(1): 149.

BACKGROUND: Lung cancer is the leading cause of cancer related deaths and its incidence is highly correlated with cigarette smoking. Nicotine, the addictive component of tobacco smoke, cannot initiate tumors, but can promote proliferation, migration, and invasion of cells in vitro and promote tumor growth and metastasis in vivo. This nicotine-mediated tumor promotion is facilitated through the activation of nicotinic acetylcholine receptors (nAChRs), specifically the alpha7 subunit. More recently, nicotine has been implicated in promoting self-renewal of stem-like side-population cells from lung cancers. This subpopulation of cancer stem-like cells has been implicated in tumor initiation, generation of the heterogeneous tumor population, metastasis, dormancy, and drug resistance. Here we describe the molecular events driving nicotine and e-cigarette extract mediated stimulation of self-renewal of stem-like cells from non-small cell lung cancer. METHODS: Experiments were conducted using A549 and H1650 non-small cell lung cancer cell lines and human mesenchymal stem cells according to protocols described in this paper. 2 muM nicotine or e-cigarette extracts was used in all relevant experiments. Biochemical analysis using western blotting, transient transfections, RT-PCR and cell biological analysis using double immunofluorescence and confocal microscopy, as well as proximity ligation assays were conducted. RESULTS: Here we demonstrate that nicotine can induce the expression of embryonic stem cell factor Sox2, which is indispensable for self-renewal and maintenance of stem cell properties in non-small cell lung adenocarcinoma (NSCLC) cells. We further demonstrate that this occurs through a nAChR-Yap1-E2F1 signaling axis downstream of Src and Yes kinases. Our data suggests Oct4 may also play a role in this process. Over the past few years, electronic cigarettes (e-cigarettes) have been promoted as healthier alternatives to traditional cigarette smoking as they do not contain tobacco; however, they do still contain nicotine. Hence we have investigated whether e-cigarette extracts can enhance tumor promoting properties similar to nicotine; we find that they can induce expression of Sox2 as well as mesenchymal markers and enhance migration and stemness of NSCLC cells. CONCLUSIONS: Our findings shed light on novel molecular mechanisms underlying the pathophysiology of smoking-related lung cancer in the context of cancer stem cell populations, and reveal new pathways involved that could potentially be exploited therapeutically.

Schrock, A., et al. (2013). "Sex determining region Y-box 2 (SOX2) amplification is an independent indicator of disease recurrence in sinonasal cancer." PLoS One**8**(3): e59201.

OBJECTIVES: The transcription factor SOX2 (3q26.3-q27) is an embryonic stem cell factor contributing to the induction of pluripotency in terminally differentiated somatic cells. Recently, amplification of the SOX2 gene locus has been described in squamous cell carcinoma (SCC) of different organ sites. Aim of this study was to investigate amplification and expression status of SOX2 in sinonasal carcinomas and to correlate the results with clinico-pathological data. MATERIALS AND METHODS: A total of 119 primary tumor samples from the sinonasal region were assessed by fluorescence in-situ hybridization and immunohistochemistry for SOX2 gene amplification and protein expression, respectively. Of these, 59 were SSCs, 18 sinonasal undifferentiated carcinomas (SNUC), 10 carcinomas associated with an inverted papilloma (INVC), 19 adenocarcinomas (AD) and 13 adenoid cystic carcinomas (ACC). RESULTS: SOX2 amplifications were found in subsets of SCCs (37.5%), SNUCs (35.3%), INVCs (37.5%) and ADs (8.3%) but not in ACCs. SOX2 amplification resulted in increased protein expression. Patients with SOX2-amplified sinonasal carcinomas showed a significantly higher rate of tumor recurrences than SOX2 non-amplified tumors. CONCLUSION: This is the first study assessing SOX2 amplification and expression in a

large cohort of sinonasal carcinomas. As opposed to AD and ACC, SOX2 amplifications were detected in more than 1/3 of all SCCs, SNUCs and INVCs. We therefore suggest that SNUCs are molecularly closely related to SCCs and INVCs and that these entities represent a subgroup of sinonasal carcinomas relying on SOX2 acquisition during oncogenesis. SOX2 amplification appears to identify sinonasal carcinomas that are more likely to relapse after primary therapy, suggesting that these patients might benefit from a more aggressive therapy regime.

Seo, E. J., et al. (2016). "Hypoxia-NOTCH1-SOX2 signaling is important for maintaining cancer stem cells in ovarian cancer." Oncotarget**7**(34): 55624-55638.

Hypoxia and NOTCH signaling have been reported to be associated with the self-renewal and drug resistance of cancer stem cells (CSCs). However, the molecular mechanisms by which hypoxia and NOTCH signaling stimulate the self-renewal and drug resistance of ovarian CSCs are poorly understood. In the present study, we identified SOX2 as a key transcription factor for CSC-like characteristics in the downstream of hypoxia-induced NOTCH signaling in epithelial ovarian cancer cells. Hypoxic treatment or overexpression of intracellular domain of NOTCH1 (NICD1) in ovarian cancer cells increased sphere formation, drug resistance, and expression of CSC-associated genes such as SOX2, ALDH, and ABC transporters. Hypoxic treatment increased the expression of NICD1, and hypoxic treatment or NICD1 overexpression increased SOX2 promoter activity, which was inhibited by deletion of HIF-1 or CSL binding sites. Furthermore, DAPT treatment decreased the effect of hypoxic treatment, and SOX2 knockdown decreased the effect of hypoxic treatment and NICD overexpression on sphere formation and drug resistance in established ovarian cancer cell lines and primary ovarian cancer cells. These results suggest that hypoxia-NOTCH1-SOX2 signaling axis is important for activation of ovarian CSCs, which may provide a novel opportunity for developing therapeutics to eradicate CSCs in ovarian cancer patients.

Shih, J., et al. (2014). "Dominant B-cell epitopes from cancer/stem cell antigen SOX2 recognized by serum samples from cancer patients." Am J Clin Exp Immunol**3**(2): 84-90.

Human sex determining region Y-box 2 (SOX2) is an important transcriptional factor involved in the pluripotency and stemness of human embryonic stem cells. SOX2 plays important roles in maintaining cancer stem cell activities of melanoma and cancers of the brain, prostate, breast, and lung. SOX2 is also a lineage survival oncogene for squamous cell carcinoma of the lung and esophagus. Spontaneous cellular and humoral immune responses against SOX2 present in cancer patients classify it as a tumor-associated antigen (TAA) shared by lung cancer, glioblastoma, and prostate cancer among others. In this study, B-cell epitopes were predicted using computer-assisted algorithms. Synthetic peptides based on the prediction were screened for recognition by serum samples from cancer patients using ELISA. Two dominant B-cell epitopes, SOX2:52-87 and SOX2:98-124 were identified. Prostate cancer, glioblastoma and lung cancer serum samples that recognized the above SOX2 epitopes also recognized the full-length protein based on Western blot. These B-cell epitopes may be used in assessing humoral immune responses against SOX2 in cancer immunotherapy and stem cell-related transplantation.

Shima, H., et al. (2016). "SOX2 and ALDH1 as Predictors of Operable Breast Cancer."Anticancer Res**36**(6): 2945-2953.

AIM: Sex-determining region Y-box binding protein-2 (SOX2) and aldehyde dehydrogenase-1 (ALDH1) are known cancer stem-cell markers, and represent candidate predictors for breast cancer prognosis. In this study we investigated the relationships between SOX2/ALDH1 expression and prognosis. MATERIALS AND METHODS: One hunred and two breast cancer surgical specimens were immunohistochemically analyzed for SOX2 and ALDH1 expression. RESULTS: Disease-free survival (DFS) and overall survival (OS) were significantly poorer for SOX2-positive patients than SOX2-negative (p=0.0024 and p=0.0021, respectively), and for ALDH1-positive patients than ALDH1-negative (p=0.0049 and p=0.0083). DFS and OS were worse for SOX2- or ALDH1-positive patients than double-negative (p=0.0053 and p=0.0166). While an obvious tendency toward worse DFS was seen for estrogen receptor (ER)-negative patients, and attenuated for ER-positive, only SOX2/ALDH1 any-positive patients showed significantly poorer DFS (p=0.0258). CONCLUSION: SOX2 and ALDH1 can be considered markers of poor prognosis, particularly in ER-negative patients. SOX2/ALDH1 any-positivity might also offer a reliable predictor of poor prognosis.

Siegle, J. M., et al. (2014). "SOX2 is a cancer-specific regulator of tumour initiating potential in cutaneous squamous cell carcinoma." Nat Commun**5**: 4511.

Although the principles that balance stem cell self-renewal and differentiation in normal tissue homeostasis are beginning to emerge, it is still unclear whether cancer cells with tumour initiating potential

are similarly governed, or whether they have acquired distinct mechanisms to sustain self-renewal and long-term tumour growth. Here we show that the transcription factor Sox2, which is not expressed in normal skin epithelium and is dispensable for epidermal homeostasis, marks tumour initiating cells (TICs) in cutaneous squamous cell carcinomas (SCCs). We demonstrate that Sox2 is required for SCC growth in mouse and human, where it enhances Nrp1/Vegf signalling to promote the expansion of TICs along the tumour-stroma interface. Our findings suggest that distinct transcriptional programmes govern self-renewal and long-term growth of TICs and normal skin epithelial stem and progenitor cells. These programmes present promising diagnostic markers and targets for cancer-specific therapies.

Singh, S., et al. (2012). "EGFR/Src/Akt signaling modulates Sox2 expression and self-renewal of stem-like side-population cells in non-small cell lung cancer." Mol Cancer**11**: 73.

BACKGROUND: Cancer stem cells are thought to be responsible for the initiation and progression of cancers. In non-small cell lung cancers (NSCLCs), Hoechst 33342 dye effluxing side population (SP) cells are shown to have stem cell like properties. The oncogenic capacity of cancer stem-like cells is in part due to their ability to self-renew; however the mechanistic correlation between oncogenic pathways and self-renewal of cancer stem-like cells has remained elusive. Here we characterized the SP cells at the molecular level and evaluated its ability to generate tumors at the orthotopic site in the lung microenvironment. Further, we investigated if the self-renewal of SP cells is dependent on EGFR mediated signaling. RESULTS: SP cells were detected and isolated from multiple NSCLC cell lines (H1650, H1975, A549), as well as primary human tumor explants grown in nude mice. SP cells demonstrated stem-like properties including ability to self-renew and grow as spheres; they were able to generate primary and metastatic tumors upon orthotopic implantation into the lung of SCID mice. In vitro study revealed elevated expression of stem cell associated markers like Oct4, Sox2 and Nanog as well as demonstrated intrinsic epithelial to mesenchymal transition features in SP cells. Further, we show that abrogation of EGFR, Src and Akt signaling through pharmacological or genetic inhibitors suppresses the self-renewal growth and expansion of SP-cells and resulted in specific downregulation of Sox2 protein expression. siRNA mediated depletion of Sox2 significantly blocked the SP phenotype as well as its self-renewal capacity; whereas other transcription factors like Oct4 and Nanog played a relatively lesser role in regulating self-renewal. Interestingly, Sox2 was elevated in metastatic foci of human NSCLC samples. CONCLUSIONS: Our findings suggest that Sox2 is a novel target of EGFR-Src-Akt signaling in NSCLCs that modulates self-renewal and expansion of stem-like cells from NSCLC. Therefore, the outcome of the EGFR-Src-Akt targeted therapy may rely upon the expression and function of Sox2 within the NSCLC-CSCs.

Singh, S. K., et al. (2015). "Antithetical NFATc1-Sox2 and p53-miR200 signaling networks govern pancreatic cancer cell plasticity." EMBO J**34**(4): 517-530.

In adaptation to oncogenic signals, pancreatic ductal adenocarcinoma (PDAC) cells undergo epithelial-mesenchymal transition (EMT), a process combining tumor cell dedifferentiation with acquisition of stemness features. However, the mechanisms linking oncogene-induced signaling pathways with EMT and stemness remain largely elusive. Here, we uncover the inflammation-induced transcription factor NFATc1 as a central regulator of pancreatic cancer cell plasticity. In particular, we show that NFATc1 drives EMT reprogramming and maintains pancreatic cancer cells in a stem cell-like state through Sox2-dependent transcription of EMT and stemness factors. Intriguingly, NFATc1-Sox2 complex-mediated PDAC dedifferentiation and progression is opposed by antithetical p53-miR200c signaling, and inactivation of the tumor suppressor pathway is essential for tumor dedifferentiation and dissemination both in genetically engineered mouse models (GEMM) and human PDAC. Based on these findings, we propose the existence of a hierarchical signaling network regulating PDAC cell plasticity and suggest that the molecular decision between epithelial cell preservation and conversion into a dedifferentiated cancer stem cell-like phenotype depends on opposing levels of p53 and NFATc1 signaling activities.

Skoda, J., et al. (2016). "Cancer stem cell markers in pediatric sarcomas: Sox2 is associated with tumorigenicity in immunodeficient mice." Tumour Biol**37**(7): 9535-9548.

The three most frequent pediatric sarcomas, i.e., Ewing's sarcoma, osteosarcoma, and rhabdomyosarcoma, were examined in this study: three cell lines derived from three primary tumor samples were analyzed from each of these tumor types. Detailed comparative analysis of the expression of three putative cancer stem cell markers related to sarcomas-ABCG2, CD133, and nestin-was performed on both primary tumor tissues and corresponding cell lines. The obtained results showed that the frequency of ABCG2-positive and CD133-positive cells was predominantly increased in the respective cell lines but

that the high levels of nestin expression were reduced in both osteosarcomas and rhabdomyosarcomas under in vitro conditions. These findings suggest the selection advantage of cells expressing ABCG2 or CD133, but the functional tests in NOD/SCID gamma mice did not confirm the tumorigenic potential of cells harboring this phenotype. Subsequent analysis of the expression of common stem cell markers revealed an evident relationship between the expression of the transcription factor Sox2 and the tumorigenicity of the cell lines in immunodeficient mice: the Sox2 levels were highest in the two cell lines that were demonstrated as tumorigenic. Furthermore, Sox2-positive cells were found in the respective primary tumors and all xenograft tumors showed apparent accumulation of these cells. All of these findings support our conclusion that regardless of the expression of ABCG2, CD133 and nestin, only cells displaying increased Sox2 expression are directly involved in tumor initiation and growth; therefore, these cells fit the definition of the cancer stem cell phenotype.

Sodja, E., et al. (2016). "The prognostic value of whole blood SOX2, NANOG and OCT4 mRNA expression in advanced small-cell lung cancer."Radiol Oncol**50**(2): 188-196.

BACKGROUND: The data on expression and clinical impact of cancer stem cell markers SOX2, NANOG and OCT4 in lung cancer is still lacking. The aim of our study was to compare SOX2, NANOG and OCT4 mRNA expression levels in whole blood between advanced small-cell lung cancer (SCLC) patients and healthy controls, and to correlate mRNA expression with progression-free survival (PFS) after first-line chemotherapy and overall survival (OS) in advanced SCLC patients. PATIENTS AND METHODS: 50 advanced SCLC patients treated with standard chemotherapy and followed at University Clinic Golnik, Slovenia, between 2009 and 2013 were prospectively included. SOX2, NANOG and OCT4 mRNA expression levels were determined using TaqMan qPCR in whole blood collected prior to chemotherapy. Whole blood of 34 matched healthy individuals with no cancerous disease was also tested. RESULTS: SOX2 mRNA expression was significantly higher in whole blood of SCLC patients compared to healthy controls ($p = 0.006$). Significant correlation between SOX2 mRNA expression levels and the number of distant metastatic sites was established ($p = 0.027$). In survival analysis, patients with high SOX2 expression had shorter OS ($p = 0.017$) and PFS ($p = 0.046$). In multivariate Cox analysis, an independent value of high SOX2 expression for shorter OS ($p = 0.002$), but not PFS was confirmed. No significant differences were observed for NANOG or OCT4 expression levels when comparing SCLC patients and healthy controls neither when analysing survival outcomes in SCLC patients. CONCLUSIONS: SOX2 mRNA expression in whole blood might be a promising non-invasive marker for molecular screening of SCLC and important prognostic marker in advanced chemotherapy-treated SCLC patients, altogether indicating important role of cancer stem-like cell (CSC) regulators in cancer spread. Further evaluation of SOX2 as a possible screening/prognostic marker and a therapeutic target of SCLC is warranted.

Srinivasan, D., et al. (2018). "Androgen receptor expression reduces stemness characteristics of prostate cancer cells (PC3) by repression of CD44 and SOX2." J Cell Biochem.

Studies have shown that a subgroup of tumor cells possess stemness characteristics having self-renewal capacity and the ability to form new tumors. We sought to identify the plausible stemness factor that determines the "molecular signature" of prostate cancer (PCa) cells derived from different metastases (PC3, PCa2b, LNCaP, and DU145) and whether androgen receptor (AR) influences the maintenance of stemness features. Here we show sex-determining region Y (SRY)-box 2 (SOX2) as a putative stem cell marker in PC3 PCa cells and not in DU145, PCa2b, or LNCaP cells. PCa2b and PC3 cells were derived from bone metastases. PCa2b cells which are positive for the AR failed to demonstrate the expression of either cluster of differentiation 44 (CD44) or SOX2. Knockdown (KD) of AR in these cells did not affect the expression of either CD44 or SOX2. Conversely, PC3 cells, which are negative for AR, expressed both CD44 and SOX2. However, the expression of AR downregulated the expression of both CD44 and SOX2 in PC3 cells. CD44 regulates SOX2 expression as KD of CD44 and reduces SOX2 levels considerably. SOX2 KD attenuated not only the expression of SNAIL and SLUG but also the migration and tumorsphere formation in PC3 cells. Collectively, our findings underscore a novel role of CD44 signaling in the maintenance of stemness and progression of cancer through SOX2 in AR-independent PC3 cells. SOX2 has a role in the regulation of expression of SNAIL and SLUG. SOX2 could be a potential therapeutic target to thwart the progression of SOX2-positive cancer cells or recurrence of androgen-independent PCa.

Stolzenburg, S., et al. (2012)."Targeted silencing of the oncogenic transcription factor SOX2 in breast cancer."Nucleic Acids Res**40**(14): 6725-6740.

The transcription factor (TF) SOX2 is essential for the maintenance of pluripotency and self-renewal in embryonic stem cells. In addition to its normal stem

cell function, SOX2 over-expression is associated with cancer development. The ability to selectively target this and other oncogenic TFs in cells, however, remains a significant challenge due to the 'undruggable' characteristics of these molecules. Here, we employ a zinc finger (ZF)-based artificial TF (ATF) approach to selectively suppress SOX2 gene expression in cancer cells. We engineered four different proteins each composed of 6ZF arrays designed to bind 18 bp sites in the SOX2 promoter and enhancer region, which controls SOX2 methylation. The 6ZF domains were linked to the Kruppel Associated Box (SKD) repressor domain. Three engineered proteins were able to bind their endogenous target sites and effectively suppress SOX2 expression (up to 95% repression efficiencies) in breast cancer cells. Targeted down-regulation of SOX2 expression resulted in decreased tumor cell proliferation and colony formation in these cells. Furthermore, induced expression of an ATF in a mouse model inhibited breast cancer cell growth. Collectively, these findings demonstrate the effectiveness and therapeutic potential of engineered ATFs to mediate potent and long-lasting down-regulation of oncogenic TF expression in cancer cells.

Takeda, K., et al. (2018). "Sox2 is associated with cancer stem-like properties in colorectal cancer." Sci Rep**8**(1): 17639.

Sox2 is known as the undifferentiated cell marker. Recent studies have shown that Sox2 may also be involved in the maintenance of cancer stem cells (CSCs) in skin and bladder cancers. In this study, we aimed to clarify the role of Sox2 in colorectal CSCs. Sox2 expression was measured in colon cancer cells and colorectal clinical samples by qRT-PCR and western blot analysis. To visualize the active Sox2 mRNA production, we generated a Sox2 promoter-dependent DsRed fluorescence emission system. Colon cancer cell lines and colorectal tumor tissues generally expressed the Sox2 protein. Knockdown of Sox2 by siRNA led to increased proliferative activity in Caco2 cells. Kaplan-Meier survival curves showed that the group with high Sox2 mRNA expression had a worse prognosis for relapse-free survival (RFS) than the low expression group (P = 0.045, median follow-up 60.0 months). Time-lapse image analysis revealed that most DsRed (+) cells exhibited typical asymmetric cell division and had higher CSC marker expressions. The DsRed (+) cells exhibited chemoresistance and they grew slower in vitro, yet they established rather larger tumors in vivo. Our data suggest that Sox2 may be a potential biomarker for colorectal CSCs.

Tam, W. L. and H. H. Ng (2014). "Sox2: masterminding the root of cancer." Cancer Cell**26**(1): 3-5.

The transcription factor Sox2 is a master regulator that maintains stemness in embryonic stem cells and neural stem cells. Using elegant lineage tracing strategies and genetic reporter mouse models, two studies (one of which is by Vanner and colleagues in this issue of Cancer Cell) now demonstrate that rare Sox2-expressing cells are the founding cancer stem cell population driving tumor initiation and therapy resistance.

Tian, T., et al. (2012). "Sox2 enhances the tumorigenicity and chemoresistance of cancer stem-like cells derived from gastric cancer." J Biomed Res**26**(5): 336-345.

Gastric cancer stem-like cells (GCSCs) have been identified to possess the ability of self-renewal and tumor initiation. However, the mechanisms involved remain largely unknown. Here, we isolated and characterized the GCSCs by side population (SP) sorting procedure and cultured sphere cells (SC) from human gastric cancer cell lines SGC-7901, BGC-823, MGC-803, HGC-27 and MKN-28. The sorting and culture assay revealed that SP cells proliferated in an asymmetric division manner. In addition, SP cells exhibited a higher potential of spheroid colony formation and greater drug resistance than non-SP cells (NSP). Moreover, the SC were found with enhanced capabilities of drug resistance in vitro and tumorigenicity in vivo. Sox2 mRNA and protein was highly and significantly overexpressed in the SP cells and SC. Importantly, downregulation of Sox2 with siRNA obviously reduced spheroid colony formation and doxorubicin efflux, as well as increased apoptosis rate in sphere cells in vitro and suppressed tumorigenicity in vivo. These results suggest that both SP cells and cultured SC enrich with GCSCs and that Sox2 plays a pivotal role in sustaining stem cell properties and might be a potential target for gastric cancer therapy.

Tian, Y., et al. (2014). "SOX2 oncogenes amplified and operate to activate AKT signaling in gastric cancer and predict immunotherapy responsiveness." J Cancer Res Clin Oncol**140**(7): 1117-1124.

INTRODUCTION: Gastric cancer is the second leading cause of cancer mortality in the world. Whether the oncogene, amplified on chromosome 3q26, SOX2, a master transcriptional regulator of stemness, operate to drive strong growth phenotype in gastric cancer were unknown. MATERIALS AND METHODS: The gene expression changes of SOX2 in human gastric cancer tissues compared with non-

cancerous tissues was detected using real-time quantitative reverse transcriptase-polymerase chain reaction (QRT-PCR) analysis and immunohistochemistry, which identified the gene overexpression of SOX2 in gastric cancer. Moreover, we discovered that SOX2 promoted cancer cell proliferation in vitro/vivo and SOX2 expression correlated with elevated AKT phosphorylation in gastric cancer, while the AKT phosphorylation was required for SOX2's oncogenic effects. Next, our data point to the usefulness of SOX2 overexpression, as a new predictive marker for responsiveness to trastuzumab. CONCLUSION: SOX2 is a commonly activated tumor promoter that activate AKT signaling in gastric cancer and a new predictive marker for targeted therapy.

Toschi, L., et al. (2014). "Increased SOX2 gene copy number is associated with FGFR1 and PIK3CA gene gain in non-small cell lung cancer and predicts improved survival in early stage disease." PLoS One**9**(4): e95303.

BACKGROUND: We aimed to investigate prevalence and prognostic role of SOX2, PIK3CA, FGFR1 and BRF2 gene gain in patients with surgically resected non-small cell lung cancer (NSCLC). METHODS: SOX2, PIK3CA, FGFR1 and BRF2 gene copy number was assessed by fluorescence in situ hybridization (FISH) in arrayed tissue cores from 447 resected NSCLCs. RESULTS: Increased gene copy number (FISH+) for SOX2, PIK3CA, FGFR1 and BRF2 was observed in 23.6%, 29.2%, 16.6% and 14.9% of cases, respectively. FISH+ status for each gene was significantly associated with smoking history, squamous cell carcinoma (SCC) histology, and increased copy number of the other studied genes. Multivariate analysis of overall survival indicated increased SOX2 gene copy number (P = 0.008), stage I-II (P<0.001), and adenocarcinoma or SCC histology (P = 0.016) as independent, favorable prognostic factors. A statistically significant interaction was observed between stage and SOX2 gene status (P = 0.021), indicating that the prognostic impact of SOX2 gene gain differs across stages and is limited to patients with stage I-II disease (HR 0.44, 95% CI: 0.25-0.77; P = 0.004, adjusted for histology). CONCLUSIONS: Increased SOX2 gene copy number is an independent and favorable prognostic factor in surgically resected, early stage NSCLC, regardless of histology. SOX2, PIK3CA, FGFR1 and BRF2 gene gains are likely to occur concurrently, with potentially relevant implications for the development of new therapeutic strategies.

Tripathi, S. C., et al. (2017). "MCAM Mediates Chemoresistance in Small-Cell Lung Cancer via the PI3K/AKT/SOX2 Signaling Pathway." Cancer Res**77**(16): 4414-4425.

Despite favorable responses to initial therapy, small-cell lung cancer (SCLC) relapse occurs within a year and exhibits resistance to multiple drugs. Because of limited accessibility of patient tissues for research purposes, SCLC patient-derived xenografts (PDX) have provided the best opportunity to address this limitation. Here, we sought to identify novel mechanisms involved in SCLC chemoresistance. Through in-depth proteomic profiling, we identified MCAM as a markedly upregulated surface receptor in chemoresistant SCLC cell lines and in chemoresistant PDX compared with matched treatment-naive tumors. MCAM depletion in chemoresistant cells reduced cell proliferation and reduced the IC50 inhibitory concentration of chemotherapeutic drugs in vitro This MCAM-mediated sensitization to chemotherapy occurred via SOX2-dependent upregulation of mitochondrial 37S ribosomal protein 1/ATP-binding cassette subfamily C member 1 (MRP1/ABCC1) and the PI3/AKT pathway. Metabolomic profiling revealed that MCAM modulated lactate production in chemoresistant cells that exhibit a distinct metabolic phenotype characterized by low oxidative phosphorylation. Our results suggest that MCAM may serve as a novel therapeutic target to overcome chemoresistance in SCLC. Cancer Res; 77(16); 4414-25. (c)2017 AACR.

Tulsyan, S., et al. (2014)."Significant association of combination of OCT4, NANOG, and SOX2 gene polymorphisms in susceptibility and response to treatment in North Indian breast cancer patients."Cancer Chemother Pharmacol**74**(5): 1065-1078.

PURPOSE: Dysregulations of regulatory genes in embryonic stem cells (ESCs) gene polymorphisms may lead to breast cancer cell growth, differentiation, and tumor metastasis. METHODS: Polymorphisms in OCT4 (rs3130932), NANOG (rs11055786), LIN28 (rs4274112), and SOX2 (rs11915160) genes were evaluated for susceptibility in 297 breast cancer females and 273 healthy controls from north Indian population. Response to neo-adjuvant chemotherapy was followed in 128 locally advanced breast cancer patients along with clinicopathological features. Genotyping was done using TaqMan allelic discrimination assays. Statistical analysis was performed using SPSS and multifactor dimensionality reduction (MDR). RESULTS: For OCT4 gene polymorphism, protective effect of genotypes AC [P corr = 0.031, OR = 0.63 (0.44-0.91)] and AC+CC [P corr = 0.031, OR = 0.68 (0.48-0.95)] was seen in patients. However, no association of NANOG, LIN28, and SOX2 gene polymorphisms was found with

overall breast cancer susceptibility. Further, significant association of AG+GG genotype [P corr = 0.021, OR = 6.08 (1.83-20.15)] and G allele [P corr = 0.021, OR = 3.07 (1.21-7.77)] of rs4274112 polymorphism was seen with positive lymph node. For OCT4, significant association of allele C was seen with patients having negative hormone receptor [P corr = 0.021, OR = 0.51 (0.29-0.90)], but no association of any of the studied polymorphisms individually was found with response to NACT. On MDR analysis, we found combination of SNPs SOX2 rs11915160, OCT4 rs3130932, and NANOG rs11055786 to be the best interaction model for predicting breast cancer risk [p for permutation test <10(-3), OR = 2.04 (1.43-2.910] and response to NACT [p for permutation test = 0.005, OR = 2.09 (1.24-3.52)]. CONCLUSION: Combination of genetic variants of ESCs gene may have a profound effect in breast cancer risk and response to NACT.

Uozaki, H., et al. (2011). "Transcriptional factor typing with SOX2, HNF4aP1, and CDX2 closely relates to tumor invasion and Epstein-Barr virus status in gastric cancer." Int J Clin Exp Pathol**4**(3): 230-240.

BACKGROUND: Gastric cancer (GC) is a major cancer, sometimes associated with Epstein-Barr virus (EBV). Some transcriptional factors (TFs) are specific to the digestive tract and related to the character of the tumors. METHODS: We studied three TFs, SOX2, CDX2, and hepatocyte nuclear factor 4 alpha-promoter 1 (HNF4aP1) in GC. First, 255 tumors including 31 EBV-associated GC were immunohistochemically examined using tissue arrays and compared TF type and mucin phenotype. We classified them into 4 TF types: N-TF type as SOX2-/HNF4aP1- tumor, G: SOX2+/HNF4aP1-, GI: SOX2+/HNF4aP1+, and I: SOX2-/HNF4aP1+. Next, 915 GCs were intensely investigated and compared with their clinicopathological factors. RESULTS: In the first study, 255 GCs were classified into N-TF 44%, G-TF 31%, GI-TF 3%, and I-TF 2%. The TF type did not strictly accord with the mucin phenotype, classified by MUC2/5AC/6/CD10 expression. EBV status was the only factor related to both the TF and mucin phenotype classifications (P<0.0001, <0.0001). TF classification is related to more factors including tumor stage, than mucin phenotype classification. The second study using 915 GCs revealed that N-TF gradually increased and I-TF decreased as GC invaded deeper. TF classification was not related to nodal involvement in each tumor stage. HNF4aP1 and CDX2 were independent factors for early stage tumor in logistic regression analysis. CONCLUSIONS: EBV-associated GC is a discriminating group in both TF and mucin phenotype. TF classification, especially the absence of HNF4aP1 and CDX2, is related to tumor invasion. TF classification is a useful marker to study the carcinogenesis of GC further.

van Schaijik, B., et al. (2018). "Subcellular localisation of the stem cell markers OCT4, SOX2, NANOG, KLF4 and c-MYC in cancer: a review." J Clin Pathol**71**(1): 88-91.

The stem cell markers octamer-binding transcription factor 4, sex-determining region Y-box 2, NANOG, Kruppel-like factor 4 and c-MYC are key factors in inducing pluripotency in somatic cells, and they have been used to detect cancer stem cell subpopulations in a range of cancer types. Recent literature has described the subcellular localisation of these markers and their potential implications on cellular function. This is a relatively complex and unexplored area of research, and the extent of the effect that subcellular localisation has on cancer development and growth is largely unknown. This review analyses this area of research in the context of the biology of stem cells and cancer and explores the potential modulating effect of subcellular localisation of these proteins as supported by the literature.

Vazquez-Martin, A., et al. (2013). "Reprogramming of non-genomic estrogen signaling by the stemness factor SOX2 enhances the tumor-initiating capacity of breast cancer cells." Cell Cycle**12**(22): 3471-3477.

The restoration of pluripotency circuits by the reactivation of endogenous stemness factors, such as SOX2, may provide a new paradigm in cancer development. The tumoral stem cell reprogramming hypothesis, i.e., the ability of stemness factors to redirect normal and differentiated tumor cells toward a less-differentiated and stem-like state, adds new layers of complexity to cancer biology, because the effects of such reprogramming may remain dormant until engaged later in response to (epi)genetic and/or (micro)environmental events. To test this hypothesis, we utilized an in vitro model of a SOX2-overexpressing cancer stem cell (CSC)-like cellular state that was recently developed in our laboratory by employing Yamanaka's nuclear reprogramming technology in the estrogen receptor alpha (ERalpha)-positive MCF-7 breast cancer cell line. Despite the acquisition of distinct molecular features that were compatible with a breast CSC-like cellular state, such as strong aldehyde dehydrogenase activity, as detected by ALDEFLUOR, and overexpression of the SSEA-4 and CD44 breast CSC markers, the tumor growth-initiating ability of SOX2-overexpressing CSC-like MCF-7 cells solely occurred in female nude mice supplemented with estradiol when compared with MCF-7 parental cells. Ser118 phosphorylation of estrogen receptor alpha (ERalpha), which is a pivotal

integrator of the genomic and nongenomic E 2/ERalpha signaling pathways, drastically accumulated in nuclear speckles in the interphase nuclei of SOX2-driven CSC-like cell populations. Moreover, SOX2-positive CSC-like cells accumulated significantly higher numbers of actively dividing cells, and the highest levels of phospho-Ser118-ERalpha occurred when chromosomes lined up on a metaphase plate. The previously unrecognized link between E 2/ERalpha signaling and SOX2-driven stem cell circuitry may significantly impact our current understanding of breast cancer initiation and progression, i.e., SOX2 can promote non-genomic E 2 signaling that leads to nuclear phospho-Ser118-ERalpha, which ultimately exacerbates genomic ER signaling in response to E 2. Because E 2 stimulation has been recently shown to enhance breast tumor-initiating cell survival by downregulating miR-140, which targets SOX2, the establishment of a bidirectional cross-talk interaction between the stem cell self-renewal regulator, SOX2, and the local and systemic ability of E 2 to increase breast CSC activity may have profound implications for the development of new CSC-directed strategies for breast cancer prevention and therapy.

Verma, N. K., et al. (2017). "Myeloid Zinc Finger 1 and GA Binding Protein Co-Operate with Sox2 in Regulating the Expression of Yes-Associated Protein 1 in Cancer Cells." Stem Cells**35**(12): 2340-2350.

The transcription factor (TF) yes-associated protein 1 (YAP1) is a major effector of the tumor suppressive Hippo signaling pathway and is also necessary to maintain pluripotency in embryonic stem cells. Elevated levels of YAP1 expression antagonize the tumor suppressive effects of the Hippo pathway that normally represses YAP1 function. High YAP1 expression is observed in several types of human cancers and is particularly prominent in cancer stem cells (CSCs). The stem cell TF Sox2, which marks and maintains CSCs in osteosarcomas (OSs), promotes YAP1 expression by binding to an intronic enhancer element and YAP1 expression is also crucial for the maintainance of OS stem cells. To further understand the regulation of YAP1 expression in OSs, we subjected the YAP1 intronic enhancer to scanning mutagenesis to identify all DNA cis-elements critical for enhancer function. Through this approach, we identified two novel TFs, GA binding protein (GABP) and myeloid zinc finger 1 (MZF1), which are essential for basal YAP1 transcription. These factors are highly expressed in OSs and bind to distinct sites in the YAP1 enhancer. Depletion of either factor leads to drastically reduced YAP1 expression and thus a reversal of stem cell properties. We also found that YAP1 can regulate the expression of Sox2 by binding to two distinct DNA binding sites upstream and downstream of the Sox2 gene. Thus, Sox2 and YAP1 reinforce each others expression to maintain stemness and tumorigenicity in OSs, but the activity of MZF1 and GABP is essential for YAP1 transcription. Stem Cells 2017;35:2340-2350.

Virant-Klun, I., et al. (2016)."Small putative NANOG, SOX2, and SSEA-4-positive stem cells resembling very small embryonic-like stem cells in sections of ovarian tissue in patients with ovarian cancer."J Ovarian Res**9**: 12.

BACKGROUND: In previous studies it has been found that in cell cultures of human adult ovaries there is a population of small stem cells with diameters of 2-4 mum, which are present mainly in the ovarian surface epithelium and are comparable to very small embryonic-like stem cells (VSELs) from bone marrow. These cells are not observed by histopathologists in the ovarian tissue due to their small size and unknown clinical significance. Because these cells express a degree of pluripotency, they might be involved in the manifestation of ovarian cancer. Therefore we studied the ovarian tissue sections in women with borderline ovarian cancer and serous ovarian carcinoma to perhaps identify the small putative stem cells in situ. METHODS: In 27 women with borderline ovarian cancer and 20 women with high-grade serous ovarian carcinoma the ovarian tissue sections were stained, per standard practice, with eosin and hematoxylin staining and on NANOG, SSEA-4 and SOX2 markers, related to pluripotency, using immunohistochemistry. We focused on the presence and localization of small putative stem cells with diameters of up to 5 mum and with the nuclei spread over nearly the full cell volume. RESULTS: In ovarian sections of both borderline ovarian cancer and serous ovarian carcinoma patients we were able to identify the presence of small round cells complying with the above criteria. Some of these small cells were NANOG-positive, were located among epithelial cells in the ovarian surface epithelium and as a single cell or groups of cells/clusters in typical "chambers", were found only in the presence of ovarian cancer and not in healthy ovaries and are comparable to those in fetal ovaries. We envision that these small cells could be related to NANOG-positive tumor-like structures and oocyte-like cells in similar "chambers" found in sections of cancerous ovaries, which could support their stemness and pluripotency. Further immunohistochemistry revealed a similar population of SSEA-4 and SOX2-positive cells. CONCLUSIONS: We may conclude that putative small stem cells expressing markers, related to pluripotency, are present in the ovarian tissue sections

of women with borderline ovarian cancer and high-grade serous ovarian carcinoma thus indicating their potential involvement in ovarian cancer.

Wang, K., et al. (2018). "FGFR1-ERK1/2-SOX2 axis promotes cell proliferation, epithelial-mesenchymal transition, and metastasis in FGFR1-amplified lung cancer." Oncogene**37**(39): 5340-5354.

Epithelial-mesenchymal transition (EMT) is an important process for cancer metastasis, drug resistance, and cancer stem cells. Activation of fibroblast growth factor receptor 1 (FGFR1) was found to promote EMT and metastasis in prostate and breast cancers, but the effects and mechanisms in lung cancer was unclear. In this study, we aimed to explore whether and how activation of FGFR1 promotes EMT and metastasis in FGFR1-amplified lung cancer. We show that activation of FGFR1 by its ligand fibroblast growth factor 2 (FGF2) promoted proliferation, EMT, migration, and invasion in FGFR1-amplified lung cancer cell lines H1581 and DMS114, whereas inhibition of FGFR1 suppressed these processes. FGFR1 activation upregulated expression of Sry-related HMG box 2 (SOX2) by downstream phosphorylated ERK1/2; moreover, the upregulation of SOX2 by autophosphorylation variant ERK2_R67S plasmid transfection was not suppressed by FGFR1 inhibitor AZD4547 or MEK/ERK inhibitor AZD6244 in vitro. And SOX2 expression was also significantly upregulated in ERK2_R67S lentivirus-transfected stable cell lines in vivo. Overexpression of SOX2 promoted cell proliferation, EMT, migration, and invasion. Importantly, activation of FGFR1 could not promote these processes in SOX2-silenced stable cell lines. In orthotopic and subcutaneous lung cancer xenograft models, inhibition of FGFR1 suppressed tumor growth, SOX2 expression, EMT, and metastasis in vivo; however, these processes caused by SOX2-overexpressing stable cell lines were not suppressed by FGFR1 inhibition. Higher expression of FGFR1 and SOX2 were positively correlated, and both were associated with shorter survival in lung cancer patients. In conclusion, our findings reveal that activation of FGFR1 promotes cell proliferation, EMT, and metastasis by the newly defined FGFR1-ERK1/2-SOX2 axis in FGFR1-amplified lung cancer.

Wang, L., et al. (2016). "Repression of TIF1gamma by SOX2 promotes TGF-beta-induced epithelial-mesenchymal transition in non-small-cell lung cancer." Oncogene**35**(7): 867-877.

TIF1gamma is a novel regulator of transforming growth factor (TGF)-beta/Smad signaling. Our previous studies show that dysregulated expression of transcriptional intermediary factor 1 gamma (TIF1gamma) and abnormal TGF-beta/Smad signaling are implicated in non-small-cell lung cancer (NSCLC) separately. However, how TIF1gamma contributes to NSCLC by controlling TGF-beta/Smad signaling is poorly understood. Here, we investigated the mechanistic role of TIF1gamma in TGF-beta-induced epithelial-mesenchymal transition (EMT), as well as a link between TIF1gamma and SOX2 in NSCLC. We show that TIF1gamma is a downstream target of SOX2 in NSCLC cells. SOX2 overexpression negatively regulated TIF1gamma promoter activity and thereby attenuated TIF1gamma mRNA and protein expression levels; SOX2 knockdown significantly enhanced TIF1gamma promoter activity and augmented TIF1gamma expression. Moreover, TIF1gamma mRNA expression was downregulated in human NSCLC tissues and negatively correlated with SOX2 protein, which was upregulated in NSCLC tissues. Importantly, knockdown of TIF1gamma or SOX2 overexpression augmented SMAD4 (human Mad (mothers against decapentaplegic)-related homologous protein 4)-dependent transcriptional responses, and enhanced TGF-beta-induced EMT and human NSCLC cell invasion; knockdown of SOX2 impaired TGF-beta-induced EMT and NSCLC cell invasion. In an in vivo model of metastasis, knockdown of TIF1gamma promotes NSCLC cell metastasis. In addition, our data suggested that TIF1gamma inhibited TGF-beta-induced EMT through competing with SMAD4 in NSCLC cells. Taken together, our findings reveal a new mechanism by which SOX2-mediated transcription repression of TIF1gamma promotes TGF-beta-induced EMT in NSCLC.

Wang, S., et al. (2015). "SOX2, a predictor of survival in gastric cancer, inhibits cell proliferation and metastasis by regulating PTEN." Cancer Lett**358**(2): 210-219.

Inconsistent results of SOX2 expression have been reported in gastric cancer (GC). Here, we demonstrated that SOX2 was progressively downregulated during GC development via immunochemistry in 755 human gastric specimens. Low SOX2 levels were associated with pathological stage and clinical outcome. Multivariate analysis indicated that SOX2 protein expression served as an independent prognostic marker for GC. Gain-and loss-of function studies showed the anti-proliferative, anti-metastatic, and pro-apoptotic effects of SOX2 in GC. PTEN was selected as SOX2 targets by cDNA microarray and ChIP-DSL, further identified by luciferase assays, EMSA and ChIP-PCR. PTEN upregulation in response to SOX2-enforced expression suppressed GC malignancy via regulating Akt dephosphorylation. PTEN inhibition reversed SOX2-induced anticancer effects. Moreover, concordant

positivity of SOX2 and PTEN proteins in nontumorous tissues but lost in matched GC specimens predicted a worse patient prognosis. Thus, SOX2 proved to be a new marker for evaluating GC outcome.

Wang, X., et al. (2014). "SOX2 enhances the migration and invasion of ovarian cancer cells via Src kinase." PLoS One**9**(6): e99594.

Ovarian cancer is the leading cause of death among gynecologic cancers and is the fifth leading cause of all cancer-related deaths among women. The development of novel molecular targets is therefore important to many patients. Recently, the SRY-related transcription factor SOX2 has been widely reported to be involved in multiple pathophysiological diseases, including maintenance of stem cell characteristics and carcinogenesis. Up to now, SOX2 has been mainly shown to promote the development of cancer, although its inhibitory roles in cancer have also been reported. However, the role of SOX2 in ovarian cancer is largely unknown. In the present study, we detected the expression of SOX2 in 64 human serous ovarian carcinoma (SOC) tissues and paired corresponding metastatic specimens using immunohistochemistry. The results showed that the expression of SOX2 in primary tumors is much lower than that in the corresponding metastatic lesions. We further found that SOX2 overexpression promotes proliferation, migration and invasion, while inhibiting adhesion abilities of SOC cells. Finally, we found that SOX2 targets Src kinase, a non-receptor tyrosine kinase that regulates cell migration, invasion and adhesion in SOC cells. Together, these results suggested that Src kinase is a key molecule in SOX2-mediated migration and invasion of SOC cells.

Wang, Y., et al. (2017). "Upregulation of SOX2 activated LncRNA PVT1 expression promotes breast cancer cell growth and invasion." Biochem Biophys Res Commun**493**(1): 429-436.

Increasing evidences indicated that Long noncoding RNAs (lncRNAs) played pivotal roles during tumorigenesis in multiple types of cancers, including breast cancer. This study aimed to investigate the role and function of long noncoding RNA PVT1 in the progression of breast cancer and explore the transcription factor which contributes to the regulation of PVT1 in breast cancer. Our results indicated that the expression of PVT1 was significantly upregulated in breast cancer tissues, compared with adjacent normal tissues (ANTs). And the increasing expression of PVT1 was associated with clinical stage, lymph nodes metastasis, and overall survival in breast cancer patients. Using computational screening, a transcriptional factor binding site was found between SOX2 and PVT1 promoter and the interaction between each other was further verified by chromatin immunoprecipitation (ChIP) analysis. In addition, ectopically overexpression of SOX2 can enhance breast cancer cell proliferation and invasion, while knockdown of SOX2 or PVT1 can severely attenuate this effect both in epithelial and mesenchymal types of breast cancer cells. Further evidences confirmed that overexpression of SOX2 can promote breast cancer cell EMT process; while silencing SOX2 or PVT1 expression can undermine this effect. These data suggest that elevated expression of SOX2 can activate lncRNA PVT1 expression promoted breast cancer tumorigenesis and progression. PVT1 may be a prognostic predictive biomarker for breast cancer, and the interaction of PVT1-SOX2 could be a therapeutic target in breast cancer.

Wang, Z., et al. (2014). "miR-625 down-regulation promotes proliferation and invasion in esophageal cancer by targeting Sox2." FEBS Lett**588**(6): 915-921.

miR-625 has been reported to exhibit abnormal expression in esophageal cancer (EC), but the mechanism and functions of miR-625 in esophageal cancer remain unclear. miR-625 down-regulation and Sox2 up-regulation were validated by qRT-PCR in 158 EC samples. Low expression of miR-625 promotes cell proliferation and invasion, while high expression of miR-625 has the opposite effect. Sox2, a target gene of miR-625, was examined by luciferase assay and western blot. Our data suggest that miR-625 may regulate the biological processes of EC via controlling Sox2 expression.

Wei, B., et al. (2016). "[Methylation and demethylation of SOX2 gene promoter in BGC-823 gastric cancer cells]."Zhonghua Yi Xue Za Zhi**96**(32): 2548-2553.

OBJECTIVE: To study the significance and effect of methylation status of sex determining region Y-box 2 (SOX2) gene promoter, and to investigate the effect of demethylation on the cell proliferation and invasion in BGC-823 gastric cancer cells. METHODS: Methylation-specific PCR (MSP) was used to assess the role of 5-aza-2'-deoxycytidine (5-Aza-CdR) in the methylation of SOX2 promoter in the BGC-823 cell lines treated with different concentration of 5-Aza-CdR. We mapped the expression of SOX2 in the BGC-823 cell lines by the quantitative real-time PCR (qPCR) and Western blotting before and after treatment of 5-Aza-CdR. The survival of BGC-823 cells were detected by MTT assay. The invasion and migration of BGC-823 cells were investigated by transwell methods, and the migration of BGC-823 cells was also assessed by the scratch assay exposed to 5-Aza-CdR or vehicle control. Model of transplanted

tumor on nude mouse were used to study the anticancer effect of 5-Aza-CdR in vivo by qPCR, Western blotting and immunohistochemistry. RESULTS: The methyltransferase inhibitor 5-Aza-CdR restored the loss of SOX2 expression in BGC-823 cell lines in a dose-dependent manner. The mRNA and protein expression of SOX2 had significant difference between the gastric cancer tissues and normal gastric mucosa (mRNA levels: 22.80+/-0.36 vs 20.36+/-0.45, P<0.05; protein levels: 0.49+/-0.01 vs 0.91+/-0.28, P<0.05). It also had significant difference among the BGC-823 cell lines treated with 5-Aza-CdR of the different concentrations (0, 1 and 10 mumol/L) (mRNA levels: 22.99+/-0.42 vs 21.78+/-0.41 vs 20.51+/-0.47, P<0.05; protein levels: 0.65+/-0.19 vs 0.73+/-0.13 vs 0.83+/-0.14, P<0.05). Compared with the control group (5-Aza-CdR concentration of 0 mumol/L), the survival rates of BGC-823 cell lines were significantly decreased in treatment groups (5-Aza-CdR concentrations of 1, 10 and 20 mumol/L, all P<0.05). Restored expression of SOX2 in the BGC-823 cell lines inhibited cell proliferation, colony formation and cell migration (P<0.05). Model of transplanted tumor on nude mouse in the 5-Aza-CdR group eventually had the smaller tumor size, the lighter tumor weight and the longer survival time than these in the PBS group [(286.6+/-37.5) vs (540.7+/-42.6)mm (3,) P<0.05; (325.2+/-32.2) vs (694.7+/-36.1)mg, P<0.05; (22.5+/-1.0) vs (18.7+/-1.6) d, P<0.05]. Meanwhile, the 5-Aza-CdR group increased the SOX2 protein expression levels and immunohistochemistry scores (0.96+/-0.25 vs 0.73+/-0.15, P<0.05; 6.23+/-0.45 vs 3.76+/-0.43, P<0.05). CONCLUSIONS: The SOX2 gene promotor is in the status of methylation in the BGC-823 cell. The recovery expression of SOX2 inhibits cell proliferation, invasion and the development of transplanted tumor in nude mice through DNA methyltransferase inhibition. It could suggest a new approach for the treatment of gastric cancer.

Weina, K. and J. Utikal (2014). "SOX2 and cancer: current research and its implications in the clinic." Clin Transl Med**3**: 19.

SOX2 is a gene that encodes for a transcription factor belonging to the SOX gene family and contains a high-mobility group (HMG) domain, which permits highly specific DNA binding. Consequently, SOX2 functions as an activator or suppressor of gene transcription. SOX2 has been described as an essential embryonic stem cell gene and moreover, a necessary factor for induced cellular reprogramming. SOX2 research has only recently switched focus from embryogenesis and development to SOX2's function in disease. Particularly, the role of SOX2 in cancer pathogenesis has become of interest in the field. To date, studies have shown SOX2 to be amplified in various cancer types and affect cancer cell physiology via involvement in complicated cell signaling and protein-protein interactions. Recent reviews in this field have highlighted SOX2 in mammalian physiology, development and pathology. In this review, we comprehensively compile what is known to date about SOX2's involvement in cancer biology, focusing on the most recent findings in the fields of cellular signaling and cancer stem cells. Lastly, we underscore the role of SOX2 in the clinic and highlight new findings, which may provide novel clinical applications for SOX2 as a prognostic marker, indicator of metastasis, biomarker or potential therapeutic target in some cancer types.

Wen, Y., et al. (2017). "SOX2 is required to maintain cancer stem cells in ovarian cancer." Cancer Sci**108**(4): 719-731.

Ovarian cancer cells can form spheroids under serum-free suspension culture conditions. The spheroids, which are enriched in cancer stem cells, can result in tumor dissemination and relapse. To identify new targetable molecules in ovarian cancer spheroids, we investigated the differential expression of genes in spheroids compared with that under monolayer culture conditions by qPCR microarray. We identified that SOX2 is overexpressed in spheroids. We then proved that SOX2 expression was increased in successive spheroid generations. Besides, knockdown of SOX2 expression in SKOV3 or HO8910 ovarian cancer spheroid cells decreased spheroid formation, cell proliferation, cell migration, resistance to Cisplatin treatment, tumorigenicity, and the expression of stemness-related genes and epithelial to mesenchymal transition-related genes, whereas overexpression of SOX2 in SKOV3 or HO8910 ovarian cancer cells showed the opposite effects. In addition, we found that SOX2 expression was closely associated with chemo-resistance and poor prognosis in EOC patients. These results strongly suggest that SOX2 is required to maintain cancer stem cells in ovarian cancer. Targeting SOX2 in ovarian cancer may be therapeutically beneficial.

Wilbertz, T., et al. (2011). "SOX2 gene amplification and protein overexpression are associated with better outcome in squamous cell lung cancer." Mod Pathol**24**(7): 944-953.

The transcription factor SOX2 (3q26.3-q27) is a key regulator of foregut development and an embryonic stem cell factor cooperating during induction of pluripotency in terminally differentiated somatic cells. Recently, we found SOX2 to be amplified in a subset of squamous cell lung and esophageal cancers. The aim of this study was to

explore the prognostic role of SOX2 in a large series of squamous cell carcinomas and adenocarcinomas of the lung. A total of 891 samples from two independent population-based cohorts were assessed by fluorescence in situ hybridization and immunohistochemistry. Furthermore, we assessed for associations between SOX2 amplification/upregulation and clinicopathological features. Similar results were found in the two cohorts. Within squamous cell carcinoma cases, 8% high-level as well as 68 and 65% low-level SOX2 amplifications occurred in the two cohorts, respectively. In adenocarcinomas, no high-level amplification was found and low-level amplification occurred in 6% of the two cohorts. Within squamous cell carcinomas of one cohort, SOX2 amplification was associated with lower tumor grade, while higher levels of SOX2 expression were related to younger age, smaller tumor size, and lower probability of angiolymphatic invasion and metastasis. High SOX2 expression levels proved to be a marker for prolonged overall survival among patients with squamous cell carcinomas. In conclusion, SOX2 amplification and upregulation are frequent events in squamous cell carcinomas of the lung and are associated with indicators of favorable prognosis.

Wu, F., et al. (2013). "Sox2 suppresses the invasiveness of breast cancer cells via a mechanism that is dependent on Twist1 and the status of Sox2 transcription activity." BMC Cancer**13**: 317.

BACKGROUND: Sox2, an embryonic stem cell marker, is aberrantly expressed in a subset of breast cancer (BC). While the aberrant expression of Sox2 has been shown to significantly correlate with a number of clinicopathologic parameters in BC, its biological significance in BC is incompletely understood. METHODS: In-vitro invasion assay was used to evaluate whether the expression of Sox2 is linked to the invasiveness of MCF7 and ZR751 cells. Quantitative reverse transcriptase-polymerase chain reaction (qRT-PCR) and/or Western blots were used to assess if Sox2 modulates the expression of factors known to regulate epithelial mesenchymal transition (EMT), such as Twist1. Chromatin immunoprecipitation (ChIP) was used to assess the binding of Sox2 to the promoter region of Twist1. RESULTS: We found that siRNA knockdown of Sox2 expression significantly increased the invasiveness of MCF7 and ZR751 cells. However, when MCF7 cells were separated into two distinct subsets based on their differential responsiveness to the Sox2 reporter, the Sox2-mediated effects on invasiveness was observed only in 'reporter un-responsive' cells (RU cells) but not 'reporter responsive' cells (RR cells). Correlating with these findings, siRNA knockdown of Sox2 in RU cells, but not RR cells, dramatically increased the expression of Twist1. Accordingly, using ChIP, we found evidence that Sox2 binds to the promoter region of Twist1 in RU cells only. Lastly, siRNA knockdown of Twist1 largely abrogated the regulatory effect of Sox2 on the invasiveness in RU cells, suggesting that the observed Sox2-mediated effects are Twist1-dependent. CONCLUSION: Sox2 regulates the invasiveness of BC cells via a mechanism that is dependent on Twist1 and the transcriptional status of Sox2. Our results have further highlighted a new level of biological complexity and heterogeneity of BC cells that may carry significant clinical implications.

Wu, F., et al. (2012). "Identification of two novel phenotypically distinct breast cancer cell subsets based on Sox2 transcription activity." Cell Signal**24**(11): 1989-1998.

Sox2 (sex-determining region Y-box protein 2) is a transcription factor regulating pluripotency in embryonic stem cells. Sox2 is aberrantly expressed in breast and other cancers, though its biological significance remains widely unexplored. To understand the significance of this aberrancy, we assessed the transcription activity of Sox2 in two Sox2-expressing breast cancer cell lines, MCF7 and ZR751, using a lentiviral Sox2 GFP reporter vector. Surprisingly, Sox2 transcription activity, as measured by GFP expression encoded in a Sox2 reporter construct, was detectable only in a small subset of cells in both cell lines. Purification of GFP+ cells (cells with Sox2 activity) and GFP- cells (cells without Sox2 activity) was enriched for two phenotypically distinct cell populations in both MCF7 and ZR751 cell lines. Specifically, GFP+ cells formed significantly more colonies in methylcellulose and more mammospheres in vitro compared to GFP- cells. These phenotypic differences are directly linked to Sox2 as siRNA knockdown of Sox2 in GFP+ cells abolished these abilities. To provide a mechanistic explanation to our observations, we performed gel shift and chromatin immunoprecipitation studies; Sox2 was found to bind to its DNA binding consensus sequence and the promoters of Cyclin D1 and Nanog (two known Sox2 downstream targets) only in GFP+ cells. GFP+ cells also up-regulated CD49f, phospho-GSK3beta, and beta-catenin. In summary, we have identified two novel phenotypically distinct cell subsets in two breast cancer cell lines based on their differential Sox2 transcription activity. We demonstrate that Sox2 transcription activity, and not its protein expression alone, underlies the tumorigenicity and cancer stem cell-like phenotypes in breast cancers.

Wu, Y., et al. (2013). "Quantification of serum SOX2 DNA with FQ-PCR potentially provides a

diagnostic biomarker for lung cancer." Med Oncol**30**(4): 737.

Sex-determining region Y-box 2 (SOX2), as a subunit of transcription and reprogramming factor, plays a critical role in the development and progression of many malignancies, including lung cancer through gene amplification. In the present study, we aimed to quantify the levels of serum SOX2 DNA, analyze its diagnostic value and compare it with existing clinical parameters in lung cancer, and purpose to provide a novel tumor marker for lung cancer. Serum DNA was extracted from 94 lung cancer patients, 10 benign lung diseases, and 30 healthy volunteers, and then the levels of SOX2 DNA were quantified using real-time fluorescent quantitative polymerase chain reaction (FQ-PCR). The data were analyzed by statistical software SPSS14.0. The present results show that serum SOX2 DNA level in lung cancer group was higher compared to the levels in benign lung diseases group ($u = 102.0$, $p < 0.001$) or healthy group ($u = 140.0$, $p < 0.001$), and it was closely associated with TNM stage, histopathological type, and tumor size ($p = 0.031$, $p = 0.012$, and $p = 0.010$, respectively). However, serum SOX2 DNA levels of lung cancer patients were not associated with age, gender, smoking status, lymph node metastasis, or tumor differentiation ($p > 0.05$). ROC curve showed a sensitivity of 78.9% and a specificity of 82.5% for the ability of serum SOX2 DNA to detect lung cancer at the cutoff value of 1,078.3 copies/ul. Furthermore, we assessed the associations of serum SOX2 levels with clinical existing lung tumor markers, such as squamous cell carcinoma antigen, cytokeratin fragment 21-1, and neuron-specific enolase. The sensitivity was increased from 24.9, 66.1, and 39.1 to 84.2, 92.8, and 87.5%, respectively, by the combination of serum SOX2 DNA. Taken together, quantification of serum SOX2 DNA by FQ-PCR may serve as a novel accessory diagnostic tool for the clinical screening and detection of lung cancer.

Wuebben, E. L. and A. Rizzino (2017). "The dark side of SOX2: cancer - a comprehensive overview." Oncotarget**8**(27): 44917-44943.

The pluripotency-associated transcription factor SOX2 is essential during mammalian embryogenesis and later in life, but SOX2 expression can also be highly detrimental. Over the past 10 years, SOX2 has been shown to be expressed in at least 25 different cancers. This review provides a comprehensive overview of the roles of SOX2 in cancer and focuses on two broad topics. The first delves into the expression and function of SOX2 in cancer focusing on the connection between SOX2 levels and tumor grade as well as patient survival. As part of this discussion, we address the developing connection between SOX2 expression and tumor drug resistance. We also call attention to an under-appreciated property of SOX2, its levels in actively proliferating tumor cells appear to be optimized to maximize tumor growth - too little or too much SOX2 dramatically alters tumor growth. The second topic of this review focuses on the exquisite array of molecular mechanisms that control the expression and transcriptional activity of SOX2. In addition to its complex regulation at the transcriptional level, SOX2 expression and activity are controlled carefully by microRNAs, long non-coding RNAs, and post-translational modifications. In the Conclusion and Future Perspectives section, we point out that there are still important unanswered questions. Addressing these questions is expected to lead to new insights into the functions of SOX2 in cancer, which will help design novels strategies for more effectively treating some of the most deadly cancers.

Xiang, R., et al. (2011). "Downregulation of transcription factor SOX2 in cancer stem cells suppresses growth and metastasis of lung cancer." Br J Cancer**104**(9): 1410-1417.

BACKGROUND: The cancer stem cell hypothesis suggests that neoplastic clones are maintained exclusively by a small subpopulation of cells, which have indefinite proliferation and differentiation potentials and give rise to phenotypically diverse cancer cells. Cancer stem cells have been isolated by their ability to efflux Hoechst 33342 dye and are referred to as the 'side population' (SP). METHODS AND RESULTS: The Hoechst efflux assay was used to isolate and characterize the SP from murine D121 lung carcinoma cells. Here, we demonstrated that D121-SP cells contain cancer stem cell characteristics, that is, upregulation of the transcription factors SOX2 and Oct 4 in D121-SP cells. In addition, the migration of D121-SP was decreased, and apoptosis of D121-SP was upregulated following knocking down of SOX2 in D121 cells. Importantly, downregulation of SOX2 in D121 cells markedly suppressed their metastatic potential in syngeneic mice. CONCLUSIONS: These results suggest that the SP is an enriched source of lung tumour cells with stem cell properties and that SOX2 has an important role in maintaining stem cell properties and functions that may be a potential target for effective lung cancer therapy.

Xu, Y., et al. (2017). "Sox2 Communicates with Tregs Through CCL1 to Promote the Stemness Property of Breast Cancer Cells." Stem Cells**35**(12): 2351-2365.

As an important component of the tumor microenvironment, CD4(+) CD25(+) Tregs reduce

antitumor immunity, promote angiogenesis and metastasis in breast cancer. However, their function in regulating the "stemness" of tumor cells and the communication between Tregs and cancer stem cells (CSCs) remain elusive. Here, we disclose that the primarily cultured Tregs isolated from breast-tumor-bearing Foxp3-EGFP mouse upregulate the stemness property of breast cancer cells. Tregs increased the side-population and the Aldehyde dehydrogenase-bright population of mouse breast cancer cells, promoted their sphere formation in a paracrine manner, and enhanced the expression of stemness genes, such as Sox2 and so forth. In addition, Tregs increased tumorigenesis, metastasis, and chemoresistance of breast cancer cells. Furthermore, Sox2-overexpression tumor cells activated NF-kappaB-CCL1 signaling to recruit Tregs through reducing the binding of H3K27Me3 on promoter regions of p65 and Ccl1. These findings reveal the functional interaction between Tregs and CSCs and indicate that targeting on the communication between them is a promising strategy in breast cancer therapy. Stem Cells 2017;35:2351-2365.

Yamawaki, K., et al. (2017). "Sox2-dependent inhibition of p21 is associated with poor prognosis of endometrial cancer." Cancer Sci**108**(4): 632-640.

Sex-determining region Y-box 2 (SOX2) is an essential factor involved in the self-renewal and pluripotency of embryonic stem cells and has functions in cell survival and progression in many types of cancers. Here, we found that several endometrial cancer cell lines expressed SOX2, which was required for cell growth. Additionally, SOX2 overexpression regulated the expression of cyclin-dependent kinase inhibitor 1A (CDKN1A), and SOX2 specifically bound to p21 promoter DNA in endometrial cancer cell lines expressing SOX2. Expressions of SOX2 in endometrial cancer patients were significantly correlated with histological grade and poor prognosis. Moreover, low p21 together with high SOX2 expressions in advanced endometrial cancer patients were associated with the most unfavorable outcomes of patients. These results indicated that simultaneous measurement of SOX2 and p21 expression in endometrial cancer patients may be a useful biomarker for patient prognosis.

Yan, Z. Y. and X. C. Sun (2018). "[LincRNA-ROR functions as a ceRNA to regulate Oct4, Sox2, and Nanog expression by sponging miR-145 and its effect on biologic characteristics of colonic cancer stem cells]." Zhonghua Bing Li Xue Za Zhi**47**(4): 284-290.

Objective: To investigate the impact of lincRNA-ROR, a ceRNA by binding miR-145 on the expression of the downstream genes Oct4, Sox2 and Nanog, and related biological characteristics of colon cancer stem cells, and to elucidate the clinical significance of this molecular regulatory network. Methods: Fifty-two cases of colorectal cancer tissue and adjacent tissue were collected at Nanyang City Central Hospital and Nanyang Second Hospital, Henan Province, from 2014 to 2016. Real-time quantitative polymerase chain reaction (qPCR) was used to detect the expression of lincRNA-ROR and miR-145 in colorectal cancer tissue and isolated colon cancer cells. The correlation between the expression of lincRNA-ROR, miR-145 and the clinicopathologic features of colon cancer was performed. CD44(-)CD133(-) and CD44(+) CD133(+) cells were isolated from SW1116 by using flow cytometry. The expression of CD44, CD133, Oct4, Sox2, Nanog, lincRNA-ROR and miR-145 in cells were detected by qPCR. The relationship between lincRNA-ROR, miR-145, Oct4, Sox2 and Nanog was analyzed by bioinformatics, dual luciferase reporter assay, qPCR and Western blot. The effects of silencing lincRNA-ROR on the proliferation and chemosensitivity of colon cancer stem cells were detected by MTT, colony formation. Results: LincRNA-ROR was frequently up-regulated and inversely correlated with miR-145 down-regulation in the colon cancer specimens ($P<0.05$). LincRNA-ROR was related to tumor size, lymph node involvement and distant metastasis ($P<0.05$), and miR-145 was found related to tumor size and tumor location ($P<0.05$). CD44(+) CD133(+) cells were successfully isolated from SW1116 by flow cytometry. The levels of CD44, CD133, Oct4, Sox2, Nanog, lincRNA-ROR in CD44(+) CD133(+) cells were significantly increased, while miR-145 was decreased compared with CD44(-)CD133(-)cells ($P<0.05$). The levels of CD44, CD133, lnc-ROR in CD44(+) CD133(+) cells were significantly reduced upon cell adherence, while miR-145 was significantly increased ($P<0.05$). Bioinformatics analysis revealed that lincRNA-ROR shared miRNA response elements with core transcription factors Oct4, Sox2 and Nanog. MiR-145 significantly inhibited the expression of lincRNA-ROR, Oct4, Sox2 and Nanog. Silencing lincRNA-ROR significantly inhibited colon cancer stem cells proliferation and increased the sensitivity to chemotherapy. Conclusions: Linc-ROR functions as a key ceRNA to prevent core TFs, e. g., Oct4, Sox2, Nanog, from miR-145-mediated suppression in colon cancer stem cells and regulates cell proliferation and chemosensitivity.The data may provide insights into the pathophysiological interactions of the components of genetic networks in the development of colon cancer and may lead to new therapies in the future.

Yang, F., et al. (2013)."Elevated expression of SOX2 and FGFR1 in correlation with poor prognosis in patients with small cell lung cancer."Int J Clin Exp Pathol**6**(12): 2846-2854.

OBJECTIVES: The central issue in this study is to investigate the expression of Sex determining region Y-BOX2 (SOX2) and fibroblast growth factor receptor 1 (FGFR1), evaluate their clinicopathological variables and prognostic significance in small cell lung cancer (SCLC). METHODS: Specimens from 222 SCLC patients and 53 adjacent normal lung tissues were detected by the immunohistochemistry for SOX2 and FGFR1 expression. The relationship between the expression of both markers and survival status was determined. RESULTS: Overexpression of SOX2 and FGFR1 were revealed in SCLC tumors than in normal tissues (P<0.05). SOX2 expression was associated with clinical stage (P=0.014) and lymph node status (P=0.041). Besides, FGFR1 expression was significantly higher in ever smokers (P=0.030) and late stage SCLC (P=0.005). SOX2, FGFR1 and TNM stage were independent prognostic factors for overall survival (OS) and Recurrence-free survival (RFS) by multivariate analysis. In stage I patients, only overexpression of SOX2, but not of FGFR1, predicted poor OS (0.027) and RFS (P=0.013). According to the expression of SOX2 and FGFR1, patients were categorized into three groups. Patients with elevated expression of both markers belonged to the group with the shortest RFS (P<0.0001) and OS (P<0.0001). CONCLUSIONS: Increased expression of SOX2 and FGFR1 may be available as poor prognostic indicators in SCLC patients.

Yang, F., et al. (2018). "OCT4, SOX2, and NANOG positive expression correlates with poor differentiation, advanced disease stages, and worse overall survival in HER2(+) breast cancer patients." Onco Targets Ther**11**: 7873-7881.

Objective: This study aimed to evaluate the correlations of expression of OCT4, SOX2, and NANOG with clinicopathological features and overall survival (OS) in human epidermal growth factor receptor 2-positive (HER2(+)) breast cancer (BC) patients. Methods: One hundred and thirty-four surgical HER2(+) BC patients who received doxorubicin and cyclophosphamide followed by paclitaxel and trastuzumab adjuvant therapy were enrolled in this study. Immunofluorescence assay was used to detect OCT4, SOX2, and NANOG expressions. The median follow-up duration was 104 months, and the last follow-up date was December 31, 2017. Results: The expressions of OCT4 (P=0.001), SOX2 (P=0.003), and NANOG (P=0.005) were higher in tumor tissues compared with paired adjacent tissues. OCT4 positive expression was associated with poor pathological differentiation (P=0.028), larger tumor size (P=0.022), advanced N stage (P<0.001), and higher TNM stage (P<0.001). SOX2 positive expression was correlated with poor pathological differentiation (P=0.005), larger tumor size (P=0.013), and increased T stage (P=0.024). NANOG positive expression was associated with poor pathological differentiation (P=0.028), higher N stage (P=0.001), and elevated TNM stage (P=0.001). Kaplan-Meier curves disclosed that OCT4 (P=0.001) and NANOG (P=0.001) positive expressions were associated with worse OS, while SOX2 (P=0.058) positive expression was only numerically correlated with poor OS, but without statistical significance. Further analyses revealed that co-expression of these three biomarkers disclosed even better predictive value for shorter OS. Conclusion: OCT4, SOX2, and NANOG positive expressions correlate with poor differentiation and advanced disease stage, and OCT4 and NANOG present with predictive values for poor OS in HER2(+) BC patients.

Yang, L., et al. (2017). "Predictive Value of Stemness Factor Sox2 in Gastric Cancer Is Associated with Tumor Location and Stage." PLoS One**12**(1): e0169124.

Cancer stem cells (CSCs) are thought to be the "root" of cancer. Although stemness-related factors ALDH1A1 and Sox2 have been used as markers to identify gastric CSCs, the expression pattern and significance of these factors in gastric cancer have not been sufficiently demonstrated. In this study, the expressions of ALDH1A1 and Sox2 were detected by immunohistochemistry in 122 gastric cancer specimens. And the correlation between Sox2 or ALDH1A1 expression and clinicopathological parameters and overall survival data were analyzed. The positive rate of ALDH1A1 expression was 60%, but there was no significant difference between survival rates of ALDH1A1-positive and ALDH1A1-negative patients. Sox2 was expressed in 42% of specimens and was associated with poor prognosis of patients (P = 0.015). Stratified analysis showed that Sox2 expression correlated with shorter lifespan only in patients with cardiac gastric cancers (P = 0.002) or stage I or II gastric cancers (P = 0.002); but not in patients with non-cardiac cancers (P = 0.556) or stage III or IV gastric cancers (P = 0.121). Analysis on a database cohort validated the correlation between Sox2 expression and poor prognosis in stage II cancer. Also, expression of Sox2 was associated with lymphnode metastasis in patients with cardiac gastric cancer (P = 0.037). A multivariate analysis revealed that Sox2 was an independent prognostic factor in cardiac gastric cancer. Our results indicate that predictive value of Sox2 in gastric cancer is associated

with cardiac cancer location and with early cancer stages (I and II).

Yang, N., et al. (2014). "Overexpression of SOX2 promotes migration, invasion, and epithelial-mesenchymal transition through the Wnt/beta-catenin pathway in laryngeal cancer Hep-2 cells." Tumour Biol**35**(8): 7965-7973.

SOX2 is a high-mobility group box containing transcription factor essential for the maintenance of embryonic stem cells. Recent evidence indicates that SOX2 overexpression correlates with metastasis and poor prognosis in patients with laryngeal squamous cell cancer. To investigate how SOX2 contributes to this aggressive phenotype, we introduced the human SOX2 gene into a low SOX2-expressing human laryngeal cancer cell line Hep-2. Cell migration and invasion were determined by the Transwell assay with or without Matrigel coating. The epithelial-mesenchymal transition (EMT)-related markers were assayed by Western blot analysis or immunofluorescence. Our results showed that exogenous expression of SOX2 in Hep-2 cells substantially promoted their migratory and invasive capabilities in culture. Moreover, Hep-2 cells stably overexpressing SOX2 underwent EMT phenotype, as evidenced by mesenchymal morphology, decreased expression of epithelial marker (E-cadherin), and increased expression of mesenchymal markers (N-cadherin, vimentin, fibronectin, and alpha-smooth muscle actin). Strikingly, Western blot analysis and immunofluorescence also showed that overexpression of SOX2 resulted in substantial increase and nuclear accumulation of beta-catenin in Hep-2 cells. However, small interfering RNA targeting beta-catenin significantly attenuated the reduced expression of E-cadherin and increased cell migration and invasion abilities in SOX2-overexpressing cells, suggesting that SOX2-induced EMT process, migration, and invasion are dependent on beta-catenin activation. Taken together, our findings underscore a novel role for SOX2 in laryngeal cancer migration and invasion.

Yang, N., et al. (2014). "SOX2 promotes the migration and invasion of laryngeal cancer cells by induction of MMP-2 via the PI3K/Akt/mTOR pathway." Oncol Rep**31**(6): 2651-2659.

SOX2 is a high mobility group box containing transcription factor that has been reported to be aberrantly overexpressed in various human malignancies, including laryngeal squamous cell carcinoma (LSCC). However, the potential role of SOX2 in LSCC migration and invasion remains to be elucidated. In the present study, we generated stable transformants of human LSCC cells constitutively overexpressing SOX2 and investigated the effects of SOX2 overexpression on migration and invasion in LSCC cells as well as the possible underlying mechanisms. We found that ectopic overexpression of SOX2 in LSCC cells enhanced their migratory and invasive ability in vitro, accompanied by increased expression and activity of matrix metalloproteinase (MMP)-2. Meanwhile, SOX2-induced cell migration and invasion were significantly abrogated by a neutralizing anti-MMP-2 antibody or small interfering RNA targeting MMP-2. Furthermore, overexpression of SOX2 induced phosphorylation of Akt and mammalian target of rapamycin (mTOR), which are downstream effectors of the PI3K pathway. Finally, LY294002, an inhibitor of PI3K, also markedly abolished SOX2-induced activation of the Akt/mTOR pathway and increased cell invasion and MMP-2 expression. Taken together, we conclude that SOX2 promotes migration and invasion of laryngeal cancer cells by inducing MMP-2 via the PI3K/Akt/mTOR pathway. Our findings suggest that SOX2 may serve as a potential therapeutic target for LSCC.

Yang, N., et al. (2015). "Silencing SOX2 Expression by RNA Interference Inhibits Proliferation, Invasion and Metastasis, and Induces Apoptosis through MAP4K4/JNK Signaling Pathway in Human Laryngeal Cancer TU212 Cells." J Histochem Cytochem**63**(9): 721-733.

SRY (sex determining region Y)-box 2 (SOX2) plays an important role in tumor cell metastasis and apoptosis. Laryngeal squamous cell carcinoma (LSCC), responsible for 1.5% of all cancers, is one of the most common head and neck malignancies. Accumulating evidence shows that SOX2 is overexpressed in several human tumors, including lung cancer, esophageal carcinoma, pancreatic carcinoma, breast cancer, ovarian carcinoma and glioma. Our study aimed to investigate the silencing effects of SOX2 expression using RNA interference (RNAi) on various biological processes in laryngeal cancer TU212 cells, including proliferation, apoptosis, invasion and metastasis. We also studied the involvement of the MAPK/JNK signaling pathway in the biological effects of SOX2 siRNA in TU212 cells. We found that silencing SOX2 decreased the proliferation, migration, and invasion of TU212 cells, and induced apoptosis. This effect of silencing SOX2 could be reversed by silencing MAP4K4. Therefore, we consider SOX2 as a key regulator of the upstream MAP4K4/JNK signaling pathways that could be a potential therapeutic target in the treatment of patients with or prevention of laryngeal cancer.

Yang, Y. G., et al. (2018). "Interferon-induced transmembrane protein 1-mediated EGFR/SOX2

signaling axis is essential for progression of non-small cell lung cancer." Int J Cancer.

Emerging data indicate that interferon-induced transmembrane protein 1 (IFITM1) plays an important role in many cancers. However, it remains unclear whether IFITM1 is functionally indispensable in nonsmall cell lung cancer (NSCLC). Here, using NSCLC cell lines and patient-derived samples, we show that IFITM1 is essentially required for the progression of NSCLC in vitro and in vivo. Specifically, IFITM1 depletion resulted in a significant reduction in sphere formation, migration, and invasion of NSCLC cells in vitro; these events were inversely correlated with the ectopic expression of IFITM1. In addition, tumor development was significantly impaired in the absence of IFITM1 in vivo. Mechanistically, epidermal growth factor receptor/sex-determining region Y-box 2 (EGFR/SOX2) signaling axis was compromised in the absence of IFITM1, and the ectopic expression of SOX2 partially rescued the defects caused by IFITM1 depletion. More importantly, using 226 patient-derived samples, we demonstrate that a high level of IFITM1 expression is associated with a poor overall survival (OS) rate in adenocarcinoma but not in squamous cell carcinoma. Collectively, these data suggest that IFITM1 is a poor prognostic marker of adenocarcinoma and an attractive target to develop novel therapeutics for NSCLC.

Yao, G. D., et al. (2018). "SOX2 gene expression and its role in triple negative breast cancer tissues."J Biol Regul Homeost Agents**32**(6): 1399-1406.

The aim of this work was to study the expression of SOX2 gene in triple negative breast cancer and its role. One hundred and twenty specimens of paraffin-embedded triple negative breast cancer (TNBC) tissues were collected from Harbin Medical University Cancer Hospital, Heilongjiang, China between January 2014 and March 2018. The expression of SOX2 was detected using immunohistochemistry, and the relationship between the expression of SOX2 and clinical features was analyzed. Breast cancer cell lines (normal group, SOX2 interference group, SOX2 overexpression group) were cultured in vitro to detect the proliferation and cloning ability of the cell lines. The expression of SOX2 was related to lymph node metastasis and stage of breast cancer (P less than 0.05), but was not related to age, menopause or tumor size (P > 0.05); the expression of SOX2 in the overexpression group was significantly greater than that in the normal group after 72 hours, and no significant difference between the overexpression group and the interference group was observed. The number of clone cells with a diameter of 0.5 mm in the interference group was lower compared to the normal group, and that of the overexpression group was higher, but not significant. SOX2 is associated with the high invasiveness of breast cancer and can be used as a therapeutic target to inhibit the metastasis of cancer cells. SOX2 can promote the proliferation of breast cancer cells and affect the size of clone cells in its involvement in clone.

Ye, X., et al. (2014). "beta-Catenin, a Sox2 binding partner, regulates the DNA binding and transcriptional activity of Sox2 in breast cancer cells." Cell Signal**26**(3): 492-501.

Sox2, an embryonic stem cell marker, has been recently implicated in the pathogenesis of breast cancer (BC). Using liquid chromatography-mass spectrometry and co-immunoprecipitation, we identified beta-catenin as a Sox2 binding partner in MCF7 cells. The interaction between Sox2 and beta-catenin was substantially different between the two cell subsets separated based on their differential responsiveness to a Sox2 reporter. Specifically, while beta-catenin binds to Sox2 in the nuclear fraction of cells showing reporter-responsiveness (i.e. RR cells), this interaction was not detectable in those that were reporter-unresponsive (i.e. RU cells). In RR but not in RU cells, siRNA knockdown of beta-catenin significantly upregulated the Sox2 transcriptional activity, enhanced its DNA binding and increased the expression of its target genes. Correlating with these findings, while inhibition of beta-catenin significantly downregulated the mammosphere formation efficiency in RU cells, this treatment paradoxically increased that of RR cells. To conclude, we identified that beta-catenin is an important binding partner of Sox2 and a regulator of its transcriptional activity in a small subset of BC cells. The interaction between Sox2 and beta-catenin provides a novel mechanism underlying the functional dichotomy of BC cells, which carries potential therapeutic implications.

Yoo, Y. A., et al. (2018). "The Role of Castration-Resistant Bmi1+Sox2+ Cells in Driving Recurrence in Prostate Cancer."J Natl Cancer Inst.

Background: Recurrence following androgen-deprivation therapy is associated with adverse clinical outcomes in prostate cancer, but the cellular origins and molecular mechanisms underlying this process are poorly defined. We previously identified a population of castration-resistant luminal progenitor cells expressing Bmi1 in the normal mouse prostate that can serve as a cancer cell-of-origin. Here, we investigate the potential of Bmi1-expressing tumor cells that survive castration to initiate recurrence in vivo. Methods: We employed lineage retracing in Bmi1-CreER; R26R-confetti; Ptenf/f transgenic mice to mark and follow the fate of emerging recurrent tumor clones after castration. A tissue recombination strategy

was used to rescue transgenic mouse prostates by regeneration as grafts in immunodeficient hosts. We also used a small molecule Bmi1 inhibitor, PTC-209, to directly test the role of Bmi1 in recurrence. Results: Transgenic prostate tumors (n = 17) regressed upon castration but uniformly recurred within 3 months. Residual regressed tumor lesions exhibited a transient luminal-to-basal phenotypic switch and marked cellular heterogeneity. Additionally, in these lesions, a subpopulation of Bmi1-expressing castration-resistant tumor cells overexpressed the stem cell reprogramming factor Sox2 (mean [SD] = 41.1 [3.8]%, n = 10, P <.001). Bmi1+Sox2+ cells were quiescent (BrdU+Bmi1+Sox2+ at 3.4 [1.5]% vs BrdU+Bmi1+Sox2- at 18.8 [3.4]%, n = 10, P =.009), consistent with a cancer stem cell phenotype. By lineage retracing, we established that recurrence emerges from the Bmi1+ tumor cells in regressed tumors. Furthermore, treatment with the small molecule Bmi1 inhibitor PTC-209 reduced Bmi1+Sox2+ cells (6.1 [1.4]% PTC-209 vs 38.8 [2.3]% vehicle, n = 10, P <.001) and potently suppressed recurrence (retraced clone size = 2.6 [0.5] PTC-209 vs 15.7 [5.9] vehicle, n = 12, P =.04). Conclusions: These results illustrate the utility of lineage retracing to define the cellular origins of recurrent prostate cancer and identify Bmi1+Sox2+ cells as a source of recurrence that could be targeted therapeutically.

Yoon, H. I., et al. (2016). "Overexpression of SOX2 Is Associated with Better Overall Survival in Squamous Cell Lung Cancer Patients Treated with Adjuvant Radiotherapy." Cancer Res Treat**48**(2): 473-482.

PURPOSE: The purpose of this study is to investigate the prognostic significance of SOX2 gene amplification and expression in patients with American Joint Committee on Cancer stage III lung squamous cell carcinoma (SCC) who underwent surgery followed by adjuvant radiotherapy. MATERIALS AND METHODS: Pathological specimens were obtained from 33 patients with stage III lung SCC treated with surgery followed by adjuvant radiotherapy between 1996 and 2008. SOX2 gene amplification and protein expression were analyzed using fluorescent in situ hybridization and immunohistochemistry, respectively. Patients were divided into two groups according to their SOX2 gene amplification and protein expression status. Kaplan-Meier estimates and a Cox proportional hazards model were used to identify the prognostic factors affecting patient survival. RESULTS: The median follow-up period for surviving patients was 58 months (range, 5 to 102 months). SOX2 gene amplification was observed in 22 patients and protein overexpression in 26 patients. SOX2 overexpression showed significant association with SOX2 gene amplification (p=0.002). In multivariate analysis, SOX2 overexpression was a significant prognostic factor for overall survival (OS) (hazard ratios [HR], 0.1; 95% confidence interval [CI], 0.002 to 0.5; p=0.005) and disease-free survival (DFS) (HR, 0.15; 95% CI, 0.04 to 0.65; p=0.01). Age (HR, 0.33; 95% CI, 0.11 to 0.98; p=0.046) and total radiation dose (HR, 0.13; 95% CI, 0.02 to 0.7; p=0.02) were the independent prognostic factors for OS and DFS. Patients with SOX2 amplification did not show a longer OS (p=0.95) and DFS (p=0.48). CONCLUSION: Our data suggested that SOX2 overexpression could be used as a positive prognostic factor in patients with stage III lung SCC receiving adjuvant radiotherapy.

You, L., et al. (2018)."Correlation of Cancer Stem-Cell Markers OCT4, SOX2, and NANOG with Clinicopathological Features and Prognosis in Operative Patients with Rectal Cancer."Yonsei Med J**59**(1): 35-42.

PURPOSE: To investigate the association of cancer stem-cell markers [octamer-binding transcription factor 4 (OCT4), sex determining region Y-box 2 (SOX2), and Nanog homebox (NANOG)] expression with clinicopathological properties and overall survival (OS) in operative rectal cancer (RC) patients receiving adjuvant therapy. MATERIALS AND METHODS: 153 patients with primary RC receiving surgery were enrolled. Tumor tissue and paired adjacent normal tissue sample were collected, and OCT4, SOX2, and NANOG expressions were assessed by immunofluorescent staining. The median follow-up duration was 5.2 years, and the last follow-up date was August 2016. RESULTS: Tumor tissue OCT4 (p<0.001), SOX2 (p=0.003), and NANOG (p<0.001) expressions were higher than those in adjacent tissue. OCT4 expression was positively correlated with pathological grade (R=0.185, p=0.022), tumor size (R=0.224, p=0.005), and N stage (R=0.170, p=0.036). NANOG expression was positively associated with tumor size (R=0.169, p=0.036). Kaplan-Meier suggested that OCT4(+) was associated with worse OS compared with OCT4? (p<0.001), while no association of SOX2 (p=0.121) and NANOG expressions (p=0.195) with OS was uncovered. Compared with one or no positive marker, at least two positive markers were associated with shorter OS (p<0.001), while all three positive markers were correlated with worse OS compared with two or less positive markers (p<0.001). Multivariate Cox's analysis revealed that OCT4(+) (p<0.001) and N stage (p=0.046) were independent factors for shorter OS. CONCLUSION: Tumor tissue OCT4 expression was correlated with poor differentiation, tumor size, and N

stage, and it can serve as an independent prognostic biomarker in operative patients with RC receiving adjuvant therapy.

The above contents are the collected information from Internet and public resources to offer to the people for the convenient reading and information disseminating and sharing.

References

1. Abd El-Maqsoud, N. M. and D. M. Abd El-Rehim (2014). "Clinicopathologic implications of EpCAM and Sox2 expression in breast cancer." Clin Breast Cancer14(1): e1-9.
2. Akimoto, N., et al. (2018). "Transfection of T-Box Transcription Factor BRACHYURY and SOX2 Synergistically Promote Self-Renewal and Invasive Phenotype in Oral Cancer Cells." Int J Mol Sci19(11).
3. Alqahtani, H., et al. (2016). "DDX17 (P72), a Sox2 binding partner, promotes stem-like features conferred by Sox2 in a small cell population in estrogen receptor-positive breast cancer." Cell Signal28(2): 42-50.
4. Amini, S., et al. (2014). "The expressions of stem cell markers: Oct4, Nanog, Sox2, nucleostemin, Bmi, Zfx, Tcl1, Tbx3, Dppa4, and Esrrb in bladder, colon, and prostate cancer, and certain cancer cell lines." Anat Cell Biol47(1): 1-11.
5. Askarian-Amiri, M. E., et al. (2014). "Emerging role of long non-coding RNA SOX2OT in SOX2 regulation in breast cancer." PLoS One9(7): e102140.
6. Atakan, S., et al. (2014). "Autologous anti-SOX2 antibody responses reflect intensity but not frequency of antigen expression in small cell lung cancer." BMC Clin Pathol14: 24.
7. Bae, K. M., et al. (2016). "Hypoxia regulates SOX2 expression to promote prostate cancer cell invasion and sphere formation." Am J Cancer Res6(5): 1078-1088.
8. Baidu. http://www.baidu.com. 2018.
9. Basu-Roy, U., et al. (2015). "Sox2 antagonizes the Hippo pathway to maintain stemness in cancer cells." Nat Commun6: 6411.
10. Belotte, J., et al. (2015). "Sox2 gene amplification significantly impacts overall survival in serous epithelial ovarian cancer." Reprod Sci22(1): 38-46.
11. Bora-Singhal, N., et al. (2015). "Gli1-Mediated Regulation of Sox2 Facilitates Self-Renewal of Stem-Like Cells and Confers Resistance to EGFR Inhibitors in Non-Small Cell Lung Cancer." Neoplasia17(7): 538-551.
12. Bornschein, J., et al. (2013). "Dysregulation of CDX1, CDX2 and SOX2 in patients with gastric cancer also affects the non-malignant mucosa." J Clin Pathol66(9): 819-822.
13. Boumahdi, S., et al. (2014). "SOX2 controls tumour initiation and cancer stem-cell functions in squamous-cell carcinoma." Nature511(7508): 246-250.
14. Bourguignon, L. Y., et al. (2012). "Hyaluronan-CD44v3 interaction with Oct4-Sox2-Nanog promotes miR-302 expression leading to self-renewal, clonal formation, and cisplatin resistance in cancer stem cells from head and neck squamous cell carcinoma." J Biol Chem287(39): 32800-32824.
15. Cai, C. F., et al. (2015). "[Expression of SOX2 in cervical intraepithelial neoplasia and cervical cancer and its clinical significance]." Nan Fang Yi Ke Da Xue Xue Bao35(1): 128-130.
16. Cai, C., et al. (2013). "[Expression of SOX2 in cervical intraepithelial neoplasia and cervical cancer and its clinical significance]." Nan Fang Yi Ke Da Xue Xue Bao33(1): 128-130.
17. Camilo, V., et al. (2014). "Immunohistochemical molecular phenotypes of gastric cancer based on SOX2 and CDX2 predict patient outcome." BMC Cancer14: 753.
18. Carina, V., et al. (2013). "Multiple pluripotent stem cell markers in human anaplastic thyroid cancer: the putative upstream role of SOX2." Thyroid23(7): 829-837.
19. Carrasco-Garcia, E., et al. (2016). "Paradoxical role of SOX2 in gastric cancer." Am J Cancer Res6(4): 701-713.
20. Chen, J., et al. (2017). "CD59 Regulation by SOX2 Is Required for Epithelial Cancer Stem Cells to Evade Complement Surveillance." Stem Cell Reports8(1): 140-151.
21. Chen, J., et al. (2018). "Downregulation of miR200c3p contributes to the resistance of breast cancer cells to paclitaxel by targeting SOX2." Oncol Rep40(6): 3821-3829.
22. Chen, S., et al. (2014). "SOX2 regulates apoptosis through MAP4K4-survivin signaling pathway in human lung cancer cells." Carcinogenesis35(3): 613-623.
23. Chen, S., et al. (2018). "Cancer-associated fibroblasts suppress SOX2-induced dysplasia in a lung squamous cancer coculture." Proc Natl Acad Sci U S A115(50): E11671-E11680.
24. Chhabra, R. (2018). "let-7i-5p, miR-181a-2-3p and EGF/PI3K/SOX2 axis coordinate to maintain cancer stem cell population in cervical cancer." Sci Rep8(1): 7840.
25. Cho, Y. Y., et al. (2013). "Autophagy and cellular senescence mediated by Sox2 suppress malignancy of cancer cells." PLoS One8(2): e57172.
26. Chou, M. Y., et al. (2015). "Sox2 expression involvement in the oncogenicity and radiochemoresistance of oral cancer stem cells." Oral Oncol51(1): 31-39.
27. Chou, Y. T., et al. (2013). "The emerging role of SOX2 in cell proliferation and survival and its

crosstalk with oncogenic signaling in lung cancer." Stem Cells31(12): 2607-2619.
28. Corominas-Faja, B., et al. (2013). "Nuclear reprogramming of luminal-like breast cancer cells generates Sox2-overexpressing cancer stem-like cellular states harboring transcriptional activation of the mTOR pathway." Cell Cycle12(18): 3109-3124.
29. Correia, L. L., et al. (2017). "SOX2 Drives Bronchial Dysplasia in a Novel Organotypic Model of Early Human Squamous Lung Cancer." Am J Respir Crit Care Med195(11): 1494-1508.
30. Cox, J. L., et al. (2013). "The SOX2-interactome in brain cancer cells identifies the requirement of MSI2 and USP9X for the growth of brain tumor cells." PLoS One8(5): e62857.
31. Das, T., et al. (2017). "Actinomycin D Down-regulates SOX2 Expression and Induces Death in Breast Cancer Stem Cells." Anticancer Res37(4): 1655-1663.
32. Dhodapkar, K. M., et al. (2013). "SOX2-specific adaptive immunity and response to immunotherapy in non-small cell lung cancer." Oncoimmunology2(7): e25205.
33. Dogan, I., et al. (2014). "SOX2 expression is an early event in a murine model of EGFR mutant lung cancer and promotes proliferation of a subset of EGFR mutant lung adenocarcinoma cell lines." Lung Cancer85(1): 1-6.
34. Dong, Z., et al. (2014). "Prognostic significance of SOX2 in head and neck cancer: a meta-analysis." Int J Clin Exp Med7(12): 5010-5020.
35. Du, J., et al. (2015). "Overexpression of Class III beta-tubulin, Sox2, and nuclear Survivin is predictive of taxane resistance in patients with stage III ovarian epithelial cancer." BMC Cancer15: 536.
36. Favaro, R., et al. (2014). "Sox2 is required to maintain cancer stem cells in a mouse model of high-grade oligodendroglioma." Cancer Res74(6): 1833-1844.
37. Finicelli, M., et al. (2014). "Expression of stemness genes in primary breast cancer tissues: the role of SOX2 as a prognostic marker for detection of early recurrence." Oncotarget5(20): 9678-9688.
38. Forno, I., et al. (2015). "Deregulation of MiR-34b/Sox2 Predicts Prostate Cancer Progression." PLoS One10(6): e0130060.
39. Gan, L., et al. (2018). "Extracellular matrix protein 1 promotes cell metastasis and glucose metabolism by inducing integrin beta4/FAK/SOX2/HIF-1alpha signaling pathway in gastric cancer." Oncogene37(6): 744-755.
40. Gayyed, M. F. and E. R. Tawfiek (2015). "Utility of SOX2 and Livin Co-Expression in the Prognosis of Bladder Cancer With Bilharzial and Non-Bilharzial Bladder Status." World J Oncol6(5): 446-455.
41. Google. http://www.google.com. 2018.
42. Gopal, K., et al. (2016). "Oxidative stress induces the acquisition of cancer stem-like phenotype in breast cancer detectable by using a Sox2 regulatory region-2 (SRR2) reporter." Oncotarget7(3): 3111-3127.
43. Guo, L., et al. (2018). "17beta-estradiol regulates the malignancy of cancer stem-like cells derived from the MCF7 cell line partially through Sox2." Oncol Lett15(3): 3790-3795.
44. Gupta, N., et al. (2018). "Phosphorylation of Sox2 at Threonine 116 is a Potential Marker to Identify a Subset of Breast Cancer Cells with High Tumorigenecity and Stem-Like Features." Cancers (Basel)10(2).
45. Han, Y. T., et al. (2016). "Physcion inhibits the metastatic potential of human colorectal cancer SW620 cells in vitro by suppressing the transcription factor SOX2." Acta Pharmacol Sin37(2): 264-275.
46. Hellner, K., et al. (2016). "Premalignant SOX2 overexpression in the fallopian tubes of ovarian cancer patients: Discovery and validation studies." EBioMedicine10: 137-149.
47. Herreros-Villanueva, M., et al. (2013). "SOX2 promotes dedifferentiation and imparts stem cell-like features to pancreatic cancer cells." Oncogenesis2: e61.
48. Hutz, K., et al. (2014). "The stem cell factor SOX2 regulates the tumorigenic potential in human gastric cancer cells." Carcinogenesis35(4): 942-950.
49. Iglesias, J. M., et al. (2014). "The Activation of the Sox2 RR2 Pluripotency Transcriptional Reporter in Human Breast Cancer Cell Lines is Dynamic and Labels Cells with Higher Tumorigenic Potential." Front Oncol4: 308.
50. Iida, H., et al. (2012). "Hypoxia induces CD133 expression in human lung cancer cells by up-regulation of OCT3/4 and SOX2." Int J Oncol40(1): 71-79.
51. Inoue, Y., et al. (2015). "Clinicopathological and Survival Analysis of Japanese Patients with Resected Non-Small-Cell Lung Cancer Harboring NKX2-1, SETDB1, MET, HER2, SOX2, FGFR1, or PIK3CA Gene Amplification." J Thorac Oncol10(11): 1590-1600.
52. Jeon, H. M., et al. (2011). "ID4 imparts chemoresistance and cancer stemness to glioma cells by derepressing miR-9*-mediated suppression of SOX2." Cancer Res71(9): 3410-3421.
53. Ji, J., et al. (2014). "[Effect of Sox2 on proliferation of cervical squamous cancer cell line SiHa]." Sichuan Da Xue Xue Bao Yi Xue Ban45(5): 785-788.
54. Jia, X., et al. (2011). "SOX2 promotes tumorigenesis and increases the anti-apoptotic property of human prostate cancer cell." J Mol Cell Biol3(4): 230-238.
55. Jia, Y., et al. (2018). "The role of GLI-SOX2 signaling axis for gemcitabine resistance in pancreatic cancer." Oncogene.
56. Jung, K., et al. (2014). "YB-1 regulates Sox2 to coordinately sustain stemness and tumorigenic

properties in a phenotypically distinct subset of breast cancer cells." BMC Cancer14: 328.
57. Kar, S., et al. (2017). "SOX2 function and Hedgehog signaling pathway are co-conspirators in promoting androgen independent prostate cancer." Biochim Biophys Acta Mol Basis Dis1863(1): 253-265.
58. Karachaliou, N., et al. (2013). "The role of SOX2 in small cell lung cancer, lung adenocarcinoma and squamous cell carcinoma of the lung." Transl Lung Cancer Res2(3): 172-179.
59. Kaufhold, S., et al. (2016). "Yin Yang 1 is associated with cancer stem cell transcription factors (SOX2, OCT4, BMI1) and clinical implication." J Exp Clin Cancer Res35: 84.
60. Keysar, S. B., et al. (2017). "Regulation of Head and Neck Squamous Cancer Stem Cells by PI3K and SOX2." J Natl Cancer Inst109(1).
61. Kim, B. W., et al. (2015). "Clinical significance of OCT4 and SOX2 protein expression in cervical cancer." BMC Cancer15: 1015.
62. Kitamura, H., et al. (2013). "Prognostic impact of the expression of ALDH1 and SOX2 in urothelial cancer of the upper urinary tract." Mod Pathol26(1): 117-124.
63. Kregel, S., et al. (2013). "Sox2 is an androgen receptor-repressed gene that promotes castration-resistant prostate cancer." PLoS One8(1): e53701.
64. Kuo, H. Y., et al. (2016). "Galectin-3 modulates the EGFR signalling-mediated regulation of Sox2 expression via c-Myc in lung cancer." Glycobiology26(2): 155-165.
65. Lee, S., et al. (2017). "Crosstalks between Raf-kinase inhibitor protein and cancer stem cell transcription factors (Oct4, KLF4, Sox2, Nanog)." Tumour Biol39(4): 1010428317692253.
66. Leis, O., et al. (2012). "Sox2 expression in breast tumours and activation in breast cancer stem cells." Oncogene31(11): 1354-1365.
67. Lundberg, I. V., et al. (2014). "SOX2 expression is regulated by BRAF and contributes to poor patient prognosis in colorectal cancer." PLoS One9(7): e101957.
68. Lundberg, I. V., et al. (2016). "SOX2 expression is associated with a cancer stem cell state and down-regulation of CDX2 in colorectal cancer." BMC Cancer16: 471.
69. Luo, J., et al. (2018). "SOX2 inhibits cell proliferation and metastasis, promotes apoptotic by downregulating CCND1 and PARP in gastric cancer." Am J Transl Res10(2): 639-647.
70. Ma H, Chen G. Stem cell. The Journal of American Science 2005;1(2):90-92.
71. Ma H, Cherng S. Eternal Life and Stem Cell. Nature and Science. 2007;5(1):81-96.
72. Ma H, Cherng S. Nature of Life. Life Science Journal 2005;2(1):7-15.
73. Ma H, Yang Y. Turritopsis nutricula. Nature and Science 2010;8(2):15-20. http://www.sciencepub.net/nature/ns0802/03_1279_hongbao_turritopsis_ns0802_15_20.pdf.
74. Ma H. The Nature of Time and Space. Nature and science 2003;1(1):1-11. Nature and science 2007;5(1):81-96.
75. Maddison, P., et al. (2010). "Autoimmunity to SOX2, clinical phenotype and survival in patients with small-cell lung cancer." Lung Cancer70(3): 335-339.
76. Maddison, P., et al. (2019). "The utility of anti-SOX2 antibodies for cancer prediction in patients with paraneoplastic neurological disorders." J Neuroimmunol326: 14-18.
77. Maehara, R., et al. (2018). "SOX2-silenced squamous cell carcinoma: a highly malignant form of esophageal cancer with SOX2 promoter hypermethylation." Mod Pathol31(1): 83-92.
78. Mamun, M. A., et al. (2018). "SOX2 in Cancer Stemness: Tumor Malignancy and Therapeutic Potentials." J Mol Cell Biol.
79. Marsland Press. http://www.sciencepub.net. 2018.
80. Matsika, A., et al. (2015). "Cancer stem cell markers in prostate cancer: an immunohistochemical study of ALDH1, SOX2 and EZH2." Pathology47(7): 622-628.
81. Maurizi, G., et al. (2018). "Sox2 is required for tumor development and cancer cell proliferation in osteosarcoma." Oncogene37(33): 4626-4632.
82. McCaughan, F., et al. (2010). "Progressive 3q amplification consistently targets SOX2 in preinvasive squamous lung cancer." Am J Respir Crit Care Med182(1): 83-91.
83. Migita, T., et al. (2017). "Epithelial-mesenchymal transition promotes SOX2 and NANOG expression in bladder cancer." Lab Invest.
84. Miller, T. J., et al. (2017). "The prognostic value of cancer stem-like cell markers SOX2 and CD133 in stage III colon cancer is modified by expression of the immune-related markers FoxP3, PD-L1 and CD3." Pathology49(7): 721-730.
85. Mollaoglu, G., et al. (2018). "The Lineage-Defining Transcription Factors SOX2 and NKX2-1 Determine Lung Cancer Cell Fate and Shape the Tumor Immune Microenvironment." Immunity49(4): 764-779 e769.
86. Mou, W., et al. (2015). "Expression of Sox2 in breast cancer cells promotes the recruitment of M2 macrophages to tumor microenvironment." Cancer Lett358(2): 115-123.
87. Mu, P., et al. (2017). "SOX2 promotes lineage plasticity and antiandrogen resistance in TP53- and RB1-deficient prostate cancer." Science355(6320): 84-88.
88. Mukherjee, P., et al. (2017). "Modulation of SOX2 expression delineates an end-point for paclitaxel-effectiveness in breast cancer stem cells." Sci Rep7(1): 9170.
89. Mukhopadhyay, A. and T. G. Oliver (2015). "Mighty mouse breakthroughs: a Sox2-driven

model for squamous cell lung cancer." Mol Cell Oncol2(2): e969651.
90. Mukhopadhyay, A., et al. (2014). "Sox2 cooperates with Lkb1 loss in a mouse model of squamous cell lung cancer." Cell Rep8(1): 40-49.
91. National Center for Biotechnology Information, U.S. National Library of Medicine. http://www.ncbi.nlm.nih.gov/pubmed. 2018.
92. Neumann, J., et al. (2011). "SOX2 expression correlates with lymph-node metastases and distant spread in right-sided colon cancer." BMC Cancer11: 518.
93. Ooki, A., et al. (2018). "Epigenetically regulated PAX6 drives cancer cells toward a stem-like state via GLI-SOX2 signaling axis in lung adenocarcinoma." Oncogene37(45): 5967-5981.
94. Ozen, M., et al. (2015). "Overexpression of miR-145-5p inhibits proliferation of prostate cancer cells and reduces SOX2 expression." Cancer Invest33(6): 251-258.
95. Park, G. B. and D. Kim (2017). "TLR5/7-mediated PI3K activation triggers epithelial-mesenchymal transition of ovarian cancer cells through WAVE3-dependent mesothelin or OCT4/SOX2 expression." Oncol Rep38(5): 3167-3176.
96. Phi, L. T. H., et al. (2018). "The anti-metastatic effect of ginsenoside Rb2 in colorectal cancer in an EGFR/SOX2-dependent manner." Cancer Med7(11): 5621-5631.
97. Picon-Ruiz, M., et al. (2016). "Interactions between Adipocytes and Breast Cancer Cells Stimulate Cytokine Production and Drive Src/Sox2/miR-302b-Mediated Malignant Progression." Cancer Res76(2): 491-504.
98. Piva, M., et al. (2014). "Sox2 promotes tamoxifen resistance in breast cancer cells." EMBO Mol Med6(1): 66-79.
99. Qin, J., et al. (2015). "DC120, a novel AKT inhibitor, preferentially suppresses nasopharyngeal carcinoma cancer stem-like cells by downregulating Sox2." Oncotarget6(9): 6944-6958.
100. Riggi, N., et al. (2010). "EWS-FLI-1 modulates miRNA145 and SOX2 expression to initiate mesenchymal stem cell reprogramming toward Ewing sarcoma cancer stem cells." Genes Dev24(9): 916-932.
101. Rodriguez-Pinilla, S. M., et al. (2007). "Sox2: a possible driver of the basal-like phenotype in sporadic breast cancer." Mod Pathol20(4): 474-481.
102. Rothenberg, S. M., et al. (2015). "Inhibition of mutant EGFR in lung cancer cells triggers SOX2-FOXO6-dependent survival pathways." Elife4.
103. Ruan, J., et al. (2013). "Predictive value of Sox2 expression in transurethral resection specimens in patients with T1 bladder cancer." Med Oncol30(1): 445.
104. Rudin, C. M., et al. (2012). "Comprehensive genomic analysis identifies SOX2 as a frequently amplified gene in small-cell lung cancer." Nat Genet44(10): 1111-1116.
105. Russo, M. V., et al. (2016). "SOX2 boosts major tumor progression genes in prostate cancer and is a functional biomarker of lymph node metastasis." Oncotarget7(11): 12372-12385.
106. Rybak, A. P. and D. Tang (2013). "SOX2 plays a critical role in EGFR-mediated self-renewal of human prostate cancer stem-like cells." Cell Signal25(12): 2734-2742.
107. Saigusa, S., et al. (2009). "Correlation of CD133, OCT4, and SOX2 in rectal cancer and their association with distant recurrence after chemoradiotherapy." Ann Surg Oncol16(12): 3488-3498.
108. Samulin Erdem, J., et al. (2016). "Mutations in TP53 increase the risk of SOX2 copy number alterations and silencing of TP53 reduces SOX2 expression in non-small cell lung cancer." BMC Cancer16: 28.
109. Schaal, C. M., et al. (2018). "Regulation of Sox2 and stemness by nicotine and electronic-cigarettes in non-small cell lung cancer." Mol Cancer17(1): 149.
110. Schrock, A., et al. (2013). "Sex determining region Y-box 2 (SOX2) amplification is an independent indicator of disease recurrence in sinonasal cancer." PLoS One8(3): e59201.
111. Seo, E. J., et al. (2016). "Hypoxia-NOTCH1-SOX2 signaling is important for maintaining cancer stem cells in ovarian cancer." Oncotarget7(34): 55624-55638.
112. Shih, J., et al. (2014). "Dominant B-cell epitopes from cancer/stem cell antigen SOX2 recognized by serum samples from cancer patients." Am J Clin Exp Immunol3(2): 84-90.
113. Shima, H., et al. (2016). "SOX2 and ALDH1 as Predictors of Operable Breast Cancer." Anticancer Res36(6): 2945-2953.
114. Siegle, J. M., et al. (2014). "SOX2 is a cancer-specific regulator of tumour initiating potential in cutaneous squamous cell carcinoma." Nat Commun5: 4511.
115. Singh, S. K., et al. (2015). "Antithetical NFATc1-Sox2 and p53-miR200 signaling networks govern pancreatic cancer cell plasticity." EMBO J34(4): 517-530.
116. Singh, S., et al. (2012). "EGFR/Src/Akt signaling modulates Sox2 expression and self-renewal of stem-like side-population cells in non-small cell lung cancer." Mol Cancer11: 73.
117. Skoda, J., et al. (2016). "Cancer stem cell markers in pediatric sarcomas: Sox2 is associated with tumorigenicity in immunodeficient mice." Tumour Biol37(7): 9535-9548.
118. Sodja, E., et al. (2016). "The prognostic value of whole blood SOX2, NANOG and OCT4 mRNA expression in advanced small-cell lung cancer." Radiol Oncol50(2): 188-196.

119. Srinivasan, D., et al. (2018). "Androgen receptor expression reduces stemness characteristics of prostate cancer cells (PC3) by repression of CD44 and SOX2." J Cell Biochem.
120. Stolzenburg, S., et al. (2012). "Targeted silencing of the oncogenic transcription factor SOX2 in breast cancer." Nucleic Acids Res40(14): 6725-6740.
121. Takeda, K., et al. (2018). "Sox2 is associated with cancer stem-like properties in colorectal cancer." Sci Rep8(1): 17639.
122. Tam, W. L. and H. H. Ng (2014). "Sox2: masterminding the root of cancer." Cancer Cell26(1): 3-5.
123. Tian, T., et al. (2012). "Sox2 enhances the tumorigenicity and chemoresistance of cancer stem-like cells derived from gastric cancer." J Biomed Res26(5): 336-345.
124. Tian, Y., et al. (2014). "SOX2 oncogenes amplified and operate to activate AKT signaling in gastric cancer and predict immunotherapy responsiveness." J Cancer Res Clin Oncol140(7): 1117-1124.
125. Toschi, L., et al. (2014). "Increased SOX2 gene copy number is associated with FGFR1 and PIK3CA gene gain in non-small cell lung cancer and predicts improved survival in early stage disease." PLoS One9(4): e95303.
126. Tripathi, S. C., et al. (2017). "MCAM Mediates Chemoresistance in Small-Cell Lung Cancer via the PI3K/AKT/SOX2 Signaling Pathway." Cancer Res77(16): 4414-4425.
127. Tulsyan, S., et al. (2014). "Significant association of combination of OCT4, NANOG, and SOX2 gene polymorphisms in susceptibility and response to treatment in North Indian breast cancer patients." Cancer Chemother Pharmacol74(5): 1065-1078.
128. Uozaki, H., et al. (2011). "Transcriptional factor typing with SOX2, HNF4aP1, and CDX2 closely relates to tumor invasion and Epstein-Barr virus status in gastric cancer." Int J Clin Exp Pathol4(3): 230-240.
129. van Schaijik, B., et al. (2018). "Subcellular localisation of the stem cell markers OCT4, SOX2, NANOG, KLF4 and c-MYC in cancer: a review." J Clin Pathol71(1): 88-91.
130. Vazquez-Martin, A., et al. (2013). "Reprogramming of non-genomic estrogen signaling by the stemness factor SOX2 enhances the tumor-initiating capacity of breast cancer cells." Cell Cycle12(22): 3471-3477.
131. Verma, N. K., et al. (2017). "Myeloid Zinc Finger 1 and GA Binding Protein Co-Operate with Sox2 in Regulating the Expression of Yes-Associated Protein 1 in Cancer Cells." Stem Cells35(12): 2340-2350.
132. Virant-Klun, I., et al. (2016). "Small putative NANOG, SOX2, and SSEA-4-positive stem cells resembling very small embryonic-like stem cells in sections of ovarian tissue in patients with ovarian cancer." J Ovarian Res9: 12.
133. Wang, K., et al. (2018). "FGFR1-ERK1/2-SOX2 axis promotes cell proliferation, epithelial-mesenchymal transition, and metastasis in FGFR1-amplified lung cancer." Oncogene37(39): 5340-5354.
134. Wang, L., et al. (2016). "Repression of TIF1gamma by SOX2 promotes TGF-beta-induced epithelial-mesenchymal transition in non-small-cell lung cancer." Oncogene35(7): 867-877.
135. Wang, S., et al. (2015). "SOX2, a predictor of survival in gastric cancer, inhibits cell proliferation and metastasis by regulating PTEN." Cancer Lett358(2): 210-219.
136. Wang, X., et al. (2014). "SOX2 enhances the migration and invasion of ovarian cancer cells via Src kinase." PLoS One9(6): e99594.
137. Wang, Y., et al. (2017). "Upregulation of SOX2 activated LncRNA PVT1 expression promotes breast cancer cell growth and invasion." Biochem Biophys Res Commun493(1): 429-436.
138. Wang, Z., et al. (2014). "miR-625 down-regulation promotes proliferation and invasion in esophageal cancer by targeting Sox2." FEBS Lett588(6): 915-921.
139. Wei, B., et al. (2016). "[Methylation and demethylation of SOX2 gene promoter in BGC-823 gastric cancer cells]." Zhonghua Yi Xue Za Zhi96(32): 2548-2553.
140. Weina, K. and J. Utikal (2014). "SOX2 and cancer: current research and its implications in the clinic." Clin Transl Med3: 19.
141. Wen, Y., et al. (2017). "SOX2 is required to maintain cancer stem cells in ovarian cancer." Cancer Sci108(4): 719-731.
142. Wikipedia. The free encyclopedia. http://en.wikipedia.org. 2018.
143. Wilbertz, T., et al. (2011). "SOX2 gene amplification and protein overexpression are associated with better outcome in squamous cell lung cancer." Mod Pathol24(7): 944-953.
144. Wu, F., et al. (2012). "Identification of two novel phenotypically distinct breast cancer cell subsets based on Sox2 transcription activity." Cell Signal24(11): 1989-1998.
145. Wu, F., et al. (2013). "Sox2 suppresses the invasiveness of breast cancer cells via a mechanism that is dependent on Twist1 and the status of Sox2 transcription activity." BMC Cancer13: 317.
146. Wu, Y., et al. (2013). "Quantification of serum SOX2 DNA with FQ-PCR potentially provides a diagnostic biomarker for lung cancer." Med Oncol30(4): 737.
147. Wuebben, E. L. and A. Rizzino (2017). "The dark side of SOX2: cancer - a comprehensive overview." Oncotarget8(27): 44917-44943.
148. Xiang, R., et al. (2011). "Downregulation of transcription factor SOX2 in cancer stem cells suppresses growth and metastasis of lung cancer." Br J Cancer104(9): 1410-1417.

149. Xu, Y., et al. (2017). "Sox2 Communicates with Tregs Through CCL1 to Promote the Stemness Property of Breast Cancer Cells." Stem Cells35(12): 2351-2365.
150. Yamawaki, K., et al. (2017). "Sox2-dependent inhibition of p21 is associated with poor prognosis of endometrial cancer." Cancer Sci108(4): 632-640.
151. Yan, Z. Y. and X. C. Sun (2018). "[LincRNA-ROR functions as a ceRNA to regulate Oct4, Sox2, and Nanog expression by sponging miR-145 and its effect on biologic characteristics of colonic cancer stem cells]." Zhonghua Bing Li Xue Za Zhi47(4): 284-290.
152. Yang, F., et al. (2013). "Elevated expression of SOX2 and FGFR1 in correlation with poor prognosis in patients with small cell lung cancer." Int J Clin Exp Pathol6(12): 2846-2854.
153. Yang, F., et al. (2018). "OCT4, SOX2, and NANOG positive expression correlates with poor differentiation, advanced disease stages, and worse overall survival in HER2(+) breast cancer patients." Onco Targets Ther11: 7873-7881.
154. Yang, L., et al. (2017). "Predictive Value of Stemness Factor Sox2 in Gastric Cancer Is Associated with Tumor Location and Stage." PLoS One12(1): e0169124.
155. Yang, N., et al. (2014). "Overexpression of SOX2 promotes migration, invasion, and epithelial-mesenchymal transition through the Wnt/beta-catenin pathway in laryngeal cancer Hep-2 cells." Tumour Biol35(8): 7965-7973.
156. Yang, N., et al. (2014). "SOX2 promotes the migration and invasion of laryngeal cancer cells by induction of MMP-2 via the PI3K/Akt/mTOR pathway." Oncol Rep31(6): 2651-2659.
157. Yang, N., et al. (2015). "Silencing SOX2 Expression by RNA Interference Inhibits Proliferation, Invasion and Metastasis, and Induces Apoptosis through MAP4K4/JNK Signaling Pathway in Human Laryngeal Cancer TU212 Cells." J Histochem Cytochem63(9): 721-733.
158. Yang, Y. G., et al. (2018). "Interferon-induced transmembrane protein 1-mediated EGFR/SOX2 signaling axis is essential for progression of non-small cell lung cancer." Int J Cancer.
159. Yao, G. D., et al. (2018). "SOX2 gene expression and its role in triple negative breast cancer tissues." J Biol Regul Homeost Agents32(6): 1399-1406.
160. Ye, X., et al. (2014). "beta-Catenin, a Sox2 binding partner, regulates the DNA binding and transcriptional activity of Sox2 in breast cancer cells." Cell Signal26(3): 492-501.
161. Yoo, Y. A., et al. (2018). "The Role of Castration-Resistant Bmi1+Sox2+ Cells in Driving Recurrence in Prostate Cancer." J Natl Cancer Inst.
162. Yoon, H. I., et al. (2016). "Overexpression of SOX2 Is Associated with Better Overall Survival in Squamous Cell Lung Cancer Patients Treated with Adjuvant Radiotherapy." Cancer Res Treat48(2): 473-482.
163. You, L., et al. (2018). "Correlation of Cancer Stem-Cell Markers OCT4, SOX2, and NANOG with Clinicopathological Features and Prognosis in Operative Patients with Rectal Cancer." Yonsei Med J59(1): 35-42.

2/25/2019