Closing down cancer

Working towards developing a new strategy in cancer therapy, Dr Isidro Sánchez-Garcia is examining oncogenes and their role in driving pretumoural cells to convert into full-blown cancer stem cells.

What are oncogenes, and how are they related to your oncology research and discoveries?

Since the origin of cancer within a particular tissue is often impossible to determine (due to the advanced stage at which patients enter clinics), knowledge about the etiology of cancer is derived from animal models that recapitulate human disease.

There is new evidence suggesting that a kind of reprogramming of cells could be a cause of human cancer. In other words, particular cancer-causing genes called ‘oncogenes’ turn on in cells and this ‘reprogrammes’ the cells to turn into cancer stem cells (CSCs). If the potential growth of cancer depends on CSCs with such oncogenes turned on, it will be important to know how to turn off the reprogramming process. Therefore, a few years ago, my research team and I initiated an ambitious hypothesis-driven research programme to study the function of oncogenes within CSCs.

Could you explain the molecular mechanisms that drive the development of cancerous tumours?

Using the BCR-ABL oncogene that is responsible for the development of chronic myeloid leukaemia as a model, we demonstrated cancer development can be established in mice by limiting oncogene expression to tumour-initiating stem cells. We further showed that CSC survival was BCR-ABL kinase independent, suggesting that curative approaches must focus on kinase-independent mechanisms of resistance. These studies showed that CSCs are not oncogene addicted (in contrast to the oncogene addiction showed by tumour-differentiated cells) and represented the first demonstration of development of CSC as a result of a reprogramming-like mechanism. Furthermore, the studies showed mutations that activate oncogenes have a driving role in the reprogramming process and may act as passenger mutations (or have a secondary, different role) thereafter. These findings challenge the accepted working model of the role of oncogenes in cancer.

Overall, our results not only highlight a previously unrecognised role for oncogenes in cancer, but also provide evidence for a previously unmodelled process for tumourigenesis in which the programming of the malignant phenotype has already taken place at the stem cell stage, thus uncovering a new role for oncogenes and the timing of cancer initiation.

The results from this study have been translated for use in humans, representing the first time that a preclinical model anticipates human CSC-therapeutic response. How could this development affect cancer patient care?

The existence of CSCs has a tremendously important therapeutic implication: Only by eliminating the CSCs can we avoid relapse and achieve a definitive cure. However, in order to do this we must be able to target CSCs specifically, discriminating them from the normal stem cells that maintain normal tissues, because therapies that kill normal stem cells would be lethal to the patient. This remains the biggest challenge for the coming years.

To what extent will your research contribute to new strategies for cancer therapy and methods for evaluating treatment efficacy?

If the potential growth of cancer depends on CSCs with oncogenes being turned on and functioning in a hands-off manner, it will be important to know how to turn off the reprogramming process. In my personal opinion, this is an important area on which to focus research efforts. The formation of tumour cells as a result of genetic reprogramming represents a new explanation of how cancer cells are formed and maintained.

The practical implications this has for the therapy of cancer are obviously enormous. What is clear is that CSC-based therapies must not replace, but rather complement, the current approaches, since the elimination of the tumour mass is also required in the initial stages of treatment.

Does this research face any particular hurdles or questions that must be addressed?

This discovery will also force us to explore and answer fundamental questions in cancer biology, such as how cells acquire and maintain their tumour differentiation states. We hypothesise that the epigenetic reprogramming properties of some oncogenes work as a new type of gene-target cell interaction in which oncogene exposure targets the epigenome to induce cancer development. To add new layers of complexity, the effect of such epigenetic reprogramming may remain dormant until engaged in response to later adult happenings, such as genetic or environmental events.

Importantly, the installation of epigenetic programmes that direct tumour-cell specific differentiation during cancer development is unidirectional. Therefore, the potential exists for a brief exposure to an environmental agent to disrupt epigenetic processes during development and reprogramme the epigenome for life. The ability to generate tumour stem cells from specific diseases and mutations in vivo has opened prospects for studying how different disease states develop from the start. If we can understand the regulation of the oncogene-target cell interaction, we will learn how to manipulate cellular states experimentally, and we could unlock the potential to provide great advances in human cancer medicine.
Researchers from the Institute of Molecular and Cell Biology of Cancer at the CSIC/University of Salamanca are discovering the roles oncogenes play in causing, and potentially one day curing, cancer.

**FEELING TIRED OR unwell, loss of appetite and high temperature; these are the symptoms a person who is in the early stages of chronic myeloid leukaemia (CML) displays. As the disease progresses, so does the severity of the symptoms, moving on to more dastardly ones such as headaches, breathlessness, enlarged lymph nodes and frequent infections.**

At the heart of CML is a mismatch between the number of underdeveloped white blood cells called granulocytes the body needs and how many it is producing. With time, these abnormal blood cells enlarge the spleen, fill the bone marrow and reduce the number of normal white blood cells, red blood cells and platelets the body produces. Eventually, the disease may lead to the patient’s death. Though scientists’ understanding of the biology of cancer and tumour cells has increased over the last 50 years, there is still a long way to go before humanity has the capacity to control the development of cancerous diseases such as CML.

Dr Isidro Sánchez-García is a scientist working to debilitate CML with the Institute of Molecular and Cell Biology of Cancer at the University of Salamanca, Spain. While his research has focused on CML, his work into targeting malignant stem cell populations to eradicate cancerous tumours may have a major impact on the concepts, therapies and methods for assessing treatment efficacy of cancer biology and development across the board.

**CONTEMPORARY CANCER RESEARCH**

To date, cancer research has mainly focused on stopping tumour cells from proliferating by blocking cancer-causing oncogene activity. These strategies take form in activities such as radiotherapy, which uses radiation to shrink tumours, and chemotherapy, which damages cancerous cells so they cannot reproduce and spread. Such treatments are based on the assumption that healthy cells, like those in the hair or blood, multiply at a much lower rate than tumour cells. For this reason, current cancer treatments often simply seem to stave off the inevitable as Sánchez-García explains: “With very few exceptions, anti-cancer treatments are targeted at the mechanisms of abnormal tumoural growth. These problems result in the eventual failure of therapy that is often accompanied by the development of drug resistance and by metastatic dissemination”.

Instead of trying to stop the proliferation of tumour cells, Sánchez-García has focused his research on discovering where tumour cells originate in the first place by looking at the role of oncogenes. However, this has been a difficult task, as determining the moment cancer develops in tissue is often impossible because patients do not enter clinics until the cancer has already advanced.

**FROM HEALTHY CELLS TO CANCER CELLS**

As a basis for work on cancer-causing oncogenes, Sánchez-García utilised a once controversial concept known as cancer stem cell (CSC) theory. According to the theory, which is quickly gaining steam in the scientific community, CSCs exist deep within the tumour. These CSCs are the ones that reproduce to maintain the tumour mass, and they replicate at a rate similar to healthy cells, potentially explaining why current cancer treatments are not sufficient for eradicating cancer.

Sánchez-García takes the approach that CSCs arise from an initial cell – the cancer cell-of-origin – in which the first genetic lesion linked with the development of the tumour takes place. The cell can be located anywhere in the physiological development pathway and does not need to have any similar physical characteristics with the final tumour cell; its genetic code just needs to be hijacked by what Sánchez-García calls an ‘oncogenic hit’. Once the cancer cell-of-origin (either a stem or differentiated cell) is ‘hit’, its epigenetic and transcriptional network is rewired to the invading cell’s type; it then becomes a self-replicating CSC – a tumoural cell with stem cell properties.

**MOUSE MODELS HOLD THE KEY**

To test his theory and elucidate if cancer is a stem cell-driven tissue, Sánchez-García and his team used the BCR-ABL oncogene responsible for the development of CML as a model. Pairing the BCR-ABL oncogene with a mouse’s Sca1 gene (the locus control region), the researchers restricted the expression of the selected, specific human cancer-associated oncogenes to the transgenic mouse’s stem cells. Sánchez-García reveals: “In transgenic mice, when the expression of BCR-ABL is restricted to the stem and progenitor cells, the Sca1-BCR-ABLp210 mice develop a CML that very closely recapitulates the main features of human disease”.

However, there is a big difference between humans and mice. “In these Sca1-BCR-ABLp210 mice, tumour initiation takes place at the stem cell or progenitor compartment, and all the leukaemic differentiated cells that form the main mass of the tumour have already switched off the expression of the oncogene. Therefore, BCR-ABL is not expressed in lineage-positive haematopoietic cells, not even the tumoural ones,” explains Sánchez-García.
around to maintain its expression at later stages in the cancer development.

These results represent the first in vivo genetic evidence of mechanistically connecting tumourigenesis and reprogramming of early progenitors. Their implication of the possibility of a reprogramming-like mechanism in cancer development is an important discovery, because it enables scientists to look for ways to shut down the harmful cells.

**NOVEL CANCER INTERVENTIONS**

Sánchez-García’s work shows how oncogenic reprogramming is a major cancer driver, and his findings mean that further investigations into the less rapidly dividing CSCs that maintain the tumour are essential to make progress to eliminate CML and other cancers. “The fact that CML development can be recapitulated in mice by limiting oncogene expression to Sca1+ cells implies that eliminating oncogene function in human patients is not going to interfere with the survival of the CSC or the formation of differentiated tumour cells,” Sánchez-García clarifies. “It suggests the oncogene is programming a (epi)genetic regulatory state in stem cells, that in some way persists during haematopoietic development, and which imposes a CML-specific tumour phenotype. This observation also applies to other cancer-initiating gene defects.”

However, there is still work to be done. A precise knowledge of the epigenetic rewiring is necessary before scientists can attempt any successful intervention, because tumour stem cell reprogramming largely relies on epigenetic modifications. Therapeutic intervention using tumour stem cell reprogramming as a target is important because unlike genetic changes, these can be erased, manipulated and reinitiated. Only then can CSC-based therapies be used to interfere with the cancer fate-inducing change and to complement current eradication approaches for CML and cancers in general.

This difference pushed the team to wonder how it is possible that CML developed efficiently in these mice, when in human cancers all leukaemic cells carry the oncogenic genetic lesions. According to Sánchez-García, the only explanation is that the BCR-ABL oncogene is actually ‘reprogramming’ the haematopoietic stem and progenitor cells through a ‘hit-and-run’ mechanism to make them malignant and deadly. Through this action, the oncogene turns genes on in stem cells but does not need to stay

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**Cancer cell biology**

- **Cancer cell-of-origin** – the cancer initiating cell and the first genetic lesion link to the cancerous tumour.
- **Cancer stem cell** – the cancer-maintaining cell. It is the cell that can regenerate and maintain the tumour.
- **Tumoural reprogramming** – a process in which the first oncogenic lesion hijacks and changes the epigenetic status of an initially healthy cell.

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**INTELLIGENCE**

**WHAT DRIVES THE CONVERSION OF A PRETUMORAL CLONE INTO A FULL-BLOWN CANCER?**

**OBJECTIVES**

To find a way to identify the molecular mechanisms that govern the development of cancer stem cells as a result of a reprogramming-like mechanism. The aim is to facilitate the investigation and discovery of new concepts in cancer biology and development and to provide the basis for the development of both a new strategy in cancer therapy and new methods for assessing treatment efficacy.

**KEY COLLABORATORS**

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