



# Cancer Stem Cell: Pathways and New approach to Cancer Genesis and Treatment

Somayeh Keshtkar\*, Ali Esmailzadeh

Central Tehran Branch, Islamic Azad University, Tehran, Iran

---

## ABSTRACT

The “cancer-stem-cell (CSCs) hypothesis” implies that tumors are composed of a heterogeneous population of cells, within which resides a small population of cancer stem cells that are responsible for the maintenance and propagation of the tumors. These cells possess the stem cell properties of self-renewal and of inhibited differentiation, but lost the controls operating in normal stem cells, or gained mutations that endowed them with tumorigenicity. The first conclusive evidence for CSCs was published in 1997 in Nature Medicine. Bonnet and Dick isolated a subpopulation of leukaemic cells that express a specific surface marker CD34, but lack the CD38 marker. The existence of leukaemic stem cells prompted further research into other types of cancer. CSCs have recently been identified in several solid tumors. The cancer stem cell (CSC) hypothesis provides an attractive cellular mechanism to account for the therapeutic refractoriness and dormant behavior exhibited by many of these tumors. Not only is finding the source of cancer cells necessary for successful treatments, but if current treatments of cancer do not properly destroy enough CSCs, the tumor will reappear. Including the possibility that the treatment of for instance, chemotherapy will leave only chemotherapy-resistant CSCs, and then the ensuing tumor will most likely also be resistant to chemotherapy. If the cancer tumor is detected early enough, enough of the tumor can be killed off and marginalized with traditional treatment. But as the tumor size increases, it becomes more and more difficult to remove the tumor without conferring resistance and leaving enough behind for the tumor to reappear. Our limited knowledge of normal stem cells, in part due to the overall paucity of experimental assays for their functional study, has made the CSC theory difficult to probe.

**KEY WORDS:** Cancer-stem-cell, tumors, leukaemic, chemotherapy

---

## INTRODUCTION

Stem cells are characterized by their capability for unlimited proliferation and their ability to self-renew by symmetric or asymmetric division. Symmetric division is the process by which stem cells increase their absolute number. In the case of asymmetric division, the stem cell divide to generate a different, more specialized daughter cell with a limited capacity for both division and/or survival. Also the second daughter cell still must remain “stem” to generate cells for growth and to substitute for the loss of cells. Another defining feature of a stem cell is to differentiate into a variety of cell types. Cancer stem cells (CSCs) are cancer cells (found within tumors or hematological cancers) that possess characteristics associated with normal stem cells, specifically the ability to give rise to all cell types found in a particular cancer sample. These cells are therefore tumorigenic (tumor-forming), perhaps in contrast to other non-tumorigenic cancer cells. CSCs may generate tumors through the stem cell processes of self-renewal and differentiation into multiple cell types. Such cells are proposed to persist in tumors as a distinct population and cause relapse and metastasis by giving rise to new tumors. Therefore, development of specific therapies targeted at CSCs holds hope for improvement of survival and quality of life of cancer patients, especially for sufferers of metastatic disease.

Existing cancer treatments were mostly developed on animal models, where therapies able to promote tumor shrinkage were deemed effective. However, animals could not provide a complete model of human disease. In particular, in mice, whose life spans do not exceed two years, tumor relapse is exceptionally difficult to study.

The efficacy of cancer treatments are, in the initial stages of testing, often measured by the amount of tumor mass they kill off. As CSCs would form a very small proportion of the tumor, this may not necessarily select for drugs that act specifically on the stem cells. The theory suggests that conventional chemotherapies kill differentiated or differentiating cells, which form the bulk of the tumor but are unable to generate new cells. A population of CSCs, which gave rise to it, could remain untouched and cause a relapse of the disease [1-6].

**Cancer stem cells:** A growing body of evidence is increasingly lending support to the concept that cancer can be modeled and interpreted as a stem-cell disease. This hypothesis is supported by a set of three experimental observations:

---

\*Corresponding Author: Somayeh Keshtkar (M.Sc.). Central Tehran Branch, Islamic Azad University, Tehran, Iran.  
Email: s\_keshtkar64@yahoo.com

- 1) Only a minority subset of cancer cells within each tumor is capable of initiating tumor growth when transplanted into immunodeficient mice;
- 2) Cancer cells endowed with tumorigenic properties are characterized by a distinctive profile of surface markers and can be differentially and reproducibly isolated from non-tumorigenic ones;
- 3) Tumors originated from purified tumorigenic cancer cells contain mixed populations of tumorigenic and non-tumorigenic ones, thus recreating the full diversity of the parent tumor's phenotypic repertoire.

The hypothesis of the cancer stem cells is not entirely new. It stemmed from the observation that not all the cells within a tumor can maintain tumor growth, and that large numbers of tumor cells are needed to transplant a tumor, even in an autologous context.

The suggestion of cancer stem cells is more plausible simply due to the fact it is a far less complicated to change to the capacity for self-renewal than it would be reactivate an immortal program in an already differentiated cell. Furthermore, stem cells live longer than differentiated cells, so they are exposed to external factors for a much longer time. The accumulation of genetic defects/mutations can also cause misregulation. Next we question the capacity for self-renewal of all tumor cells, and whether this is only due to the stem cancer cells or the presence of any other cells within the tumor tissue that might have the ability to generate a tumor. The concept that a rare population of tissue may be the cellular origin of cancer was proposed in 19th century by pathologists such as Cohnheim and Durante. These scientists observed similarities between embryonic and cancer tissue and differentiation. Their observation led to the hypothesis that resting embryonic stem cells may reside in adult tissue and that upon activation these stem cells may acquire the ability to give rise to cancer [7, 8].

In the late 1930s scientists showed that leukemia can be transmitted from one mouse to another by using a single undifferentiated leukemia cell [9]. Approximately 30 years later, using in-vitro colony formation assays, it was established that a rare subpopulation of acute myeloid leukemia (AML) possessed the ability of self-renewal and proliferation, and therefore going on to give rise to new tumors (10,11,12). In the 1970s researchers revisited this idea. The first conclusive evidence for CSCs was published in 1997 in Nature Medicine. Bonnet and Dick [1] isolated a subpopulation of leukaemic cells that express a specific surface marker CD34, but lack the CD38 marker. The authors established that the CD34<sup>+</sup>/CD38<sup>-</sup> subpopulation is capable of initiating tumors in NOD/SCID mice that is histological similar to the donor [2]. Studies that followed on from this had confirmed that there were similarities in the biology of stem cells and cancer stem cells. They proposed the idea that cancers may rise from quiescent tissue stem cells [13]. In 2001 studies were published with relevance to the steps that lead to carcinogenesis. These studies showed that the signal transduction pathways (such as Wnt and Hedgehog) play an important role in tumor genesis and how essential it is to consider the existence of stem cells and cancer stem cells regarding their roles in carcinogenesis [13, 14].

The association between cancer and stem cells and cancer was then further discussed for several decades [14,15] and it was found that stem cells and cancer stem cells have similar features concerning: unlimited proliferation, capacity for tissue specific differentiation, clonogenic potential, the capacity of self-renew and the specific differentiation. However differentiation is characteristic only for a specific cancer cell population; in some cancer cells terminal differentiation is defined, caused for example by loss of ability to proliferate, set off by external factors such as irradiation, chemical drugs. The finding that stem cells isolated from fetal tissue are able to differentiate into a variety of cell types was a very important step in proving the plasticity of somatic cells or stem cells that are present within differentiated tissues. It also led to the theory of trans differentiation. Simplified, it is the production of cells within tissues where they would not originally occur [16,17].

Opponents of the paradigm do not deny the existence of CSCs as such. Cancer cells must be capable of continuous proliferation and self-renewal in order to retain the many mutations required for carcinogenesis, and to sustain the growth of a tumor since differentiated cells cannot divide indefinitely (constrained by the Hayflick Limit). However, it is debated whether such cells represent a minority. If most cells of the tumor are endowed with stem cell properties there is no incentive to focus on a specific subpopulation. There is also debate on the cell of origin of these CSCs - whether they originate from stem cells that have lost the ability to regulate proliferation, or from more differentiated population of progenitor cells that have acquired abilities to self-renew which is related to the issue of stem cell plasticity [13, 15, 18].

In cancer research experiments, tumor cells are sometimes injected into an experimental animal to establish a tumor. Disease progression is then followed in time and novel drugs can be tested for their ability to inhibit it. However, efficient tumor formation requires thousands or tens of thousands of cells to be introduced. Classically, this has been explained by poor methodology (i.e. the tumor cells lose their viability during transfer) or the critical importance of the microenvironment, the particular biochemical surroundings of the injected cells. Supporters of the cancer stem cell paradigm argue that only a small fraction of the injected cells, the CSCs; have the potential to generate a tumor. In human acute myeloid leukemia the frequency of these cells is less than 1 in 10,000 [1]. Further evidence comes from histology, the study of tissue structure of tumors. Many tumors are very heterogeneous and contain multiple cell types native to the host organ. Heterogeneity is commonly retained by tumor metastases. This implies that the cell that produced them had the capacity to generate multiple cell types. In other words, it possessed multi-differentiate potential, a classical hallmark of stem cells. The existence of leukaemic stem cells prompted further research into other types of cancer. CSCs have recently been identified in several solid tumors, including cancers of the: Brain, Breast, Colon, Ovary, Pancreas and Prostate [1, 3, 4, 5, 6, 19, 20, 21].

## MATERIALS AND METHODS

**Definition of “Cancer Stem Cells”:** A fundamental problem in cancer research is identification of the cell type capable of initiating and sustaining growth of the neoplastic clone *in vivo*. The key to solving this riddle lies in determining whether every cell within the neoplasm has tumor-initiating capacity, or whether only a rare subset of cells – so-called ‘cancer stem cells’ (CSCs) – is responsible for maintenance of the neoplasm. The existence of CSCs was first proposed over 40 years ago, providing an explanation for the observed functional heterogeneity within tumors. However, proof of principle had to await the development of modern research tools for investigating the behavior of defined cell populations. The best evidence supporting the existence of CSCs has come from the study of hematological malignancies [22, 23].

Like their normal tissue counterparts, tumors are composed of heterogeneous populations of cells that differ in their apparent state of differentiation. Indeed, the differentiation features of a tumor, morphological and architectural, are the key parameter used in routine clinical practice by surgical pathologists to define a tumor’s primary anatomical origin. This simple observation suggests that tumors are not mere monoclonal expansions of cells but might actually be akin to “abnormal organs,” sustained by a diseased “cancer stem cell” (CSC) population, which is endowed with the ability to self-renew and undergo aberrant differentiation [13, 24].

A new wave of studies, however, has recently begun to address this concept using an innovative, purely empirical approach, based on an *in vivo* self-renewal assay. Starting from whole tumor tissues, cancer cells are purified into single-cell suspensions and subsequently fractionated in different subsets according to the expression of a specific repertoire of surface markers. Once isolated, individual cancer cell subsets are injected into appropriate hosts (in most cases orthotopic tissues of immune-deficient mouse strains), and the subsets are compared with respect to tumorigenic capacity [25].

It is important to note that, based on this approach, the term cancer stem cells represents a working definition with a purely operational significance. The term is used to indicate a tumor-initiating cell subset that can give rise to a heterogeneous progeny, similar in composition to the tissue from which it was originally isolated. In most cases, it is currently not possible to define with certainty the “genealogical” relationship between CSCs and normal stem cells of the corresponding tissues (i.e., whether CSCs originate directly from normal stem cells or the early stages of their progeny). Irrespective of the actual origins of CSCs, the identification of a CSC population establishes a functional hierarchy within a tumor tissue and encompasses both the self-renewal and differentiation hallmarks of stem cells. First developed in human myeloid leukemias, the CSC working model is today being progressively extended to several solid tumors, along with several biological and therapeutic implications [26].

## DISCUSSION

**Cancer stem cell pathways:** A normal stem cell may be transformed into a cancer stem cell through dysregulation of the proliferation and differentiation pathways controlling it. Scientists working on CSCs hope to design new drugs targeting these cellular mechanisms. The first findings in this area were made using haematopoietic stem cells (HSCs) and their transformed counterparts in leukemia, the disease whose stem cell origin is most strongly established. However, these pathways appear to be shared by stem cells of all organs.

**1) Bmi-1:** The Polycomb group transcriptional repressor Bmi-1 was discovered as a common oncogene activated in lymphoma [27] and later shown to specifically regulate HSCs [28]. The role of Bmi-1 has also been illustrated in neural stem cells [29]. The pathway appears to be active in CSCs of pediatric brain tumors [30].

**2) Notch:** The Notch pathway has been known to developmental biologists for decades. Its role in control of stem cell proliferation has now been demonstrated for several cell types including haematopoietic, neural and mammary stem cells [31]. Components of the Notch pathway have been proposed to act as oncogenes in mammary and other tumors [32].

**3) Sonic hedgehog and Wnt:** These developmental pathways are also strongly implicated as stem cell regulators. Both Sonic hedgehog (SHH) and Wnt pathways are commonly hyper activated in tumors and are required to sustain tumor growth. However, the Gli transcription factors that are regulated by SHH take their name from gliomas, where they are commonly expressed at high levels. A degree of crosstalk exists between the two pathways and their activation commonly goes hand-in-hand. This is a trend rather than a rule. For instance, in colon cancer hedgehog signaling appears to antagonise Wnt [33].

Sonic hedgehog blockers are available, such as cyclopamine. There is also a new water soluble cyclopamine that may be more effective in cancer treatment. There is also DMAPT, a water soluble derivative of parthenolide that targets AML (leukemia) stem cells, and possibly other CSCs as in myeloma or prostate cancer. A clinical trial of DMAPT is to start in England in late 2007 or 2008. Furthermore, GRN163L was recently started in trials to target myeloma stem cells. If it is possible to eliminate the cancer stem cell, then a potential cure may be achieved if there are no more CSCs to repopulate a cancer [34, 35].

**New approach to cancer genesis and treatment:** The cancer stem cell (CSC) hypothesis provides an attractive cellular mechanism to account for the therapeutic refractoriness and dormant behavior exhibited by many of these tumours. The clinical relevance of CSCs remains a fundamental issue but preliminary findings indicate that specific targeting may be possible [36].

The development of treatments that target cancer stem cells is an important objective, but the challenges are formidable. First, to design treatments that selectively eradicate cancer stem cells, it is useful to have the cognate normal stem cell or progenitor cell. This step requires the development of assays to characterize the function of normal stem cells and the means to define physical features (i.e., cell-surface antigen markers) that will permit their isolation. Without this knowledge, it is impossible to know whether a candidate drug is also cytotoxic to normal stem cells. Second, we need similar ways to describe cancer stem cells and appropriate functional assays must be validated. Third, it is critical to understand how cancer stem cells differ from normal stem cells, particularly with regard to mechanisms controlling cell survival and responses to injury. Ideally, a therapy should target pathways uniquely used by cancer stem cells to resist extrinsic insults or to maintain steady-state viability. Fourth, we must understand how therapies that effectively target the bulk of tumor cells fail to eradicate cancer stem cells. The reasons for this phenomenon may provide important clues for developing more effective and comprehensive regimens to attack both the tumor stem cells and the bulk of the disease [37].

An additional challenge in targeting cancer stem cells is to understand how the properties of stem cells make them particularly difficult to kill. Leukemia cancer stem cells reside in a largely quiescent state with regard to cell-cycle activity, like their normal counterparts [38,39]. Consequently, typical cytotoxic regimens that target rapidly dividing cells are unlikely to eradicate such cells. Selective targeting will therefore require regimens that kill cells independently of the cell cycle, or that selectively induce cycling of cancer stem cells. Another common feature of stem cells is expression of proteins associated with the efflux of xenobiotic toxins (e.g., multidrug-resistant proteins and related members of the ATP-binding cassette [ABC] transporter family). A variety of cancer cells, particularly during relapse, express such proteins, thus providing resistance to many chemotherapeutic agents [40, 41, 42, 43].

The extent to which cancer stem cells can mobilize all of the measures provided by evolutionary history to protect normal stem cells is not yet known, but this information is likely to be biologically and clinically significant.

A further concern is that normal stem cells and progenitor cells may prove to be more sensitive than cancer stem cells to the effects of chemotherapy. Normal colon stem cells, for example, can inhibit DNA repair mechanisms and thereby undergo apoptosis in response to DNA damage; this mechanism avoids the accumulation of harmful mutations. If, however, colon cancer cells evade this protective mechanism, then chemotherapy could preferentially spare those [44].

## REFERENCES

1. Bonnet, D. and Dick, J.E. 1997. "Human acute myeloid leukemia is organized as a hierarchy that originates from a primitive hematopoietic cell". *Nature Medicine*, 3(7): 730–737.
2. Matsui, W., Huff, C.A., Wang, Q., Malehorn, M.T., Barber, J., Tanhehco, Y., Smith, B.D., Civin, C.I. and Jones, R.J. 2004. "Characterization of clonogenic multiple myeloma cells". *Blood*, 103(6): 2332–2336.
3. Al-Hajj, M., Wicha, M.S., Benito-Hernandez, A., Morrison, S.J. and Clarke, M.F. 2003. "Prospective identification of tumorigenic breast cancer cells". *Proceedings of the National Academy of Sciences of the United States of America*, 100 (7): 3983–3988.
4. Zhang, S., Balch, C., Chan, M.W., Lai, H.C., Matei, D., Schilder, J.M., Yan, P.S., Huang, T.H. and Nephew, K.P. 2008. "Identification and characterization of ovarian cancer-initiating cells from primary human tumors". *Cancer Research*, 68 (11): 4311–4320.
5. Li, C., Heidt, D.G., Dalerba, P., Burant, C.F., Zhang, L., Adsay, V., Wicha, M., Clarke, M.F. and Simeone, D.M. 2007. "Identification of pancreatic cancer stem cells". *Cancer Research*, 67 (3): 1030–1037.
6. Maitland, N.J. and Collins, A.T. (2008). "Prostate cancer stem cells: a new target for therapy". *J. Clin. Oncol.*, 26 (17): 2862–2870.
7. Mačingová, Z. and Filip, S. 2008. Cancer stem cells– New approach to cancerogenesis and treatment. *Acta Medica*, 51(3): 139–144.
8. Dhom, G. 1994. The cancer cell and the connective tissue. *A historical retrospect. Pathologie*, 15: 271–278
9. Sell, S. 2004. Stem cell origin of cancer and differentiation therapy. *Crit Rev Oncol Hematol.*, 51: 1–28.
10. Moore, M.A., Williams, N., Metcalf, D. 1973. In vitro colony formation by normal and leukemic human hematopoietic cells: interaction between colony-forming and colony-stimulating cells. *J Natl Cancer Inst.*, 50: 591–602.
11. Robinson, W.A., Kurnick, J.E., Pike, B.L. 1971. Colony growth of human leukemic peripheral blood cells in vitro. *Blood.*, 38: 500–508.
12. Simonovitch, L., McCulloch, E.A. and Till, J.E. 1963. The distribution of colony forming cells among spleen colonies. *J Cell Physiol.*, 62: 327–336.
13. Reya, T., Morrison, S.J., Clarke, M.F. and Weissman, I.L. 2001. Stem cells, cancer and cancer stem cells. *Nature*, 414: 105–111.
14. Trosko, J.E., Chang, C.C. 1989. Stem cell theory of carcinogenesis. *Toxicol Lett.*, 49: 283.
15. Filip, S., Mokry, J. and English, D. 2006. Stem cell plasticity and carcinogenesis. *Neoplasma*, 53: 87–91

16. Filip, S., English, D. and Mokry, J. 2004. Issues in stem cell plasticity. *J Cell Mol Med*, 8:572–577.
17. Quesenberry, P.J., Colvin, G.A., Adebisi, M., Dooner, G., Dooner, M., Aliotta, J., Keaney, P., Luo, L., Demers, D., Peterson, A., Foster, B. and Greer, D. 2005. The stem cell continuum. *Ann NY Acad Sci*, 1044:228–235.
18. Filip, S. 2006. The phenomenon of stem cell plasticity: biological or physiological problems? *Stem Cell Dev.*, 15:753.
19. Singh, S.K., Clarke, I.D., Terasaki, M., Bonn, V.E., Hawkins, C., Squire, J., Dirks, P.B. 2003. "Identification of a cancer stem cell in human brain tumors". *Cancer Research*, 63(18): 5821–5828.
20. O'Brien, C.A., Pollett, A., Gallinger, S., Dick, J.E. 2007. "A human colon cancer cell capable of initiating tumour growth in immunodeficient mice". *Nature*, 445(7123): 106–110.
21. Lang, S.H., Frame, F., Collins, A. 2009. "Prostate cancer stem cells". *J. Pathol.*, 217(2): 299–306.
22. Bruce, W.R. and Van Der Gaag, H. 1963. A quantitative assay for the number of murine lymphoma cells capable of proliferation in vivo. *Nature*, 199: 79–80.
23. Dick, J.E. 2003. Breast cancer stem cells revealed. *Proc. Natl. Acad. Sci. U.S.A.*, 100: 3547–3549.
24. Clarke, M.F. and Fuller, M. 2006. Stem cells and cancer: two faces of Eve. *Cell*, 124:1111–1115.
25. Clarke, M.F. 2005. A self-renewal assay for cancer stem cells. *Cancer Chemother. Pharmacol.* 56(Suppl. 1):64–68
26. Piero Dalerba, Robert W. Cho, and Michael F. Clarke. *Cancer Stem Cells: Models and Concepts. Annu. Rev. Med.* 2007. 58:267–84.
27. Haupt, Y., Bath, M.L., Harris, A.W., Adams, J.M. 1993. "bmi-1 transgene induces lymphomas and collaborates with myc in tumorigenesis". *Oncogene*, 8(11): 3161–3164.
28. Park, I.K., Qian, D., Kiel, M., Becker, M.W., Pihalja, M., Weissman, I.L., Morrison, S.J., Clarke, M.F. 2003. "Bmi-1 is required for maintenance of adult self-renewing haematopoietic stem cells". *Nature*, 423(6937): 302–305.
29. Molofsky, A.V., Pardoll, R., Iwashita, T., Park, I.K., Clarke, M.F., Morrison, S.J. 2003. "Bmi-1 dependence distinguishes neural stem cell self-renewal from progenitor proliferation". *Nature* 425(6961): 962–967.
30. Hemmati, H.D., Nakano, I., Lazareff, J.A., Masterman-Smith, M., Geschwind, D.H., Bronner-Fraser, M., Kornblum, H.I. 2003. "Cancerous stem cells can arise from pediatric brain tumors". *Proceedings of the National Academy of Sciences of the United States of America* 100(25): 15178–15183.
31. Dontu, G., Jackson, K.W., McNicholas, E., Kawamura, M.J., Abdallah, W.M., Wicha, M.S. 2004. "Role of Notch signaling in cell-fate determination of human mammary stem/progenitor cells". *Breast Cancer Research: BCR*, 6(6): R605–15.
32. Diévar, A., Beaulieu, N., Jolicoeur, P. 1999. "Involvement of Notch1 in the development of mouse mammary tumors". *Oncogene* 18 (44): 5973–5981.
33. Beachy, P.A., Karhadkar, S.S., Berman, D.M. 2004. "Tissue repair and stem cell renewal in carcinogenesis". *Nature* 432 (7015): 324–331.
34. Zhou, B.P., Hung, M.C. 2005. "Wnt, hedgehog and snail: sister pathways that control by GSK-3 $\beta$  and  $\beta$ -Trcp in the regulation of metastasis". *Cell cycle (Georgetown, Tex.)*, 4(6): 772–776.
35. Akiyoshi, T., Nakamura, M., Koga, K., Nakashima, H., Yao, T., Tsuneyoshi, M., Tanaka, M., Katano, M. 2006. "Gli1, downregulated in colorectal cancers, inhibits proliferation of colon cancer cells involving Wnt signalling activation". *Gut* 55 (7): 991–999.
36. Visvader, J.E. and Lindeman, G.J. 2008. Cancer stem cells in solid tumours: accumulating evidence and unresolved questions. *Nature Reviews*, 8: 755–768.
37. Jordan, C.T. and Monica L. 2006. Guzman, Mark Noble. *Mechanisms of Disease Cancer Stem Cells. N Engl J Med.*, 355:1253–61.
38. Guan, Y., Gerhard, B., Hogge, D.E. 2003. Detection, isolation, and stimulation of quiescent primitive leukemic progenitor cells from patients with acute myeloid leukemia (AML). *Blood*, 101:3142–9
39. Holyoake, T., Jiang, X., Eaves, C. and Eaves, A. 1999. Isolation of a highly quiescent subpopulation of primitive leukemic cells in chronic myeloid leukemia. *Blood*, 94:2056–64.
40. Lowenberg, B., Sonneveld, P. 1998. Resistance to chemotherapy in acute leukemia. *Curr Opin Oncol*, 10:31–35.
41. Dean, M., Fojo, T. and Bates S. 2005. Tumour stem cells and drug resistance. *Nat Rev Cancer*, 5:275–84.
42. Gottesman, M.M., Fojo, T. and Bates, S.E. 2002. Multidrug resistance in cancer: role of ATP-dependent transporters. *Nat Rev Cancer*, 2:48–58.
43. Donnemberg, V.S., Donnemberg, A.D. 2005. Multiple drug resistance in cancer revisited: the cancer stem cell hypothesis. *J Clin Pharmacol*, 45:872–877.
44. Cairns, J. 2002. Somatic stem cells and the kinetics of mutagenesis and carcinogenesis. *Proc Natl Acad Sci USA*, 99:10567–70