Zhang, Clon Transgen 2012, 1:1
DOI: 10.4172/2168-9849.1000e101

Cloning & Transgenesis
ISSN: 2168-9849

Editorial Open Access

# From "Old" Cloning to "Young" Cellular Reprogramming: Nobel Prize 2012 Spotlighted the Stem Cell Work

**Yue Zhang***

*Research Center, CHUM, Notre Dame Hospital, Montreal, Canada*

**Abstract**

Here is a concise historical review and some perspectives on the spotlighted nuclear transplantation (cloning) and induced pluripotent stem cells (iPSC). Besides, we propose that iPSC may be hypothetically considered one exceptional case among "cancerous" cells. The argument between stochastic and elite model of iPSC is briefly discussed.

## Introduction

Mature cells can be rejuvenated to become pluripotent- the discovery for which Dr. Yamanaka and Dr. Gurdon won the 2012 Nobel Prize in medicine and physiology [1]. A few scientists can be recognised in this very special way. On the other hand, many other great scientists contributed to this field so they will not be snubbed here. Just to introduce a few names, Dr. James Thomson, who with Yamanaka won the King Faisal International Prize for Medicine in 2011 jointly for their work on stem cell therapy, is the pioneer to first isolate human embryonic stem (ES) cells in the lab back in 1998 and also showed that mature human body cells could be reversed into stem cells in Science in 2007 [2]. Dr. Ian Wilmut group successfully cloned Dolly, the first mammal cloned from an adult cell at the Roslin Institute in Scotland in 1996 [3]. Besides, a noted embryologist professor Tong Dizhou (TC Tung) produced the world's first cloned fish by inserting the DNA from a cell of a male carp into an egg from a female carp. He published the findings in a Chinese science journal in China in 1963, which was not translated into English and has seldom been reviewed in English literatures until recently [4,5]. However, the significance and influential outcomes of these discoveries are almost limitless, and there are exhilarating times to continue ahead.

## Gurdon's Cloning

Let's have a retrospective view on this year Nobel Prize in medicine and physiology spotlighted discoveries, which have already excited hundreds of scientists to enjoy their research better. In 1960s, Sir John B. Gurdon transplanted the nucleus from a mature intestinal cell into the immature cell nucleus in an egg cell of a frog. In this classic cloning experiment, some of those modified egg cells developed into a normal tadpole, so it supports that the specialization of cells is reversible [6]. His achievement has been credited as the 1st successful "cloning" in animals. Until now, cloned animals have been used to make pharmaceutical products in milk, to generate rejection-resistant animals for organ transplantation into humans, and as some preclinical models of human disease [1]. A specialized form of nuclear transfer for faulty mitochondria [7] has the potential to treat mitochondrial disease and is being developed for use in humans. However, it is his publication [6] and the follow-ups that initially ignited the controversy of ethical issue discussion later on the human cloning too.

## Yamanaka's iPSC

More than 40 years later, in order to avoid the use of surplus embryos, Yamanaka and Takahashi [8] showed in 2006 that they reprogrammed mature cells in mice to become immature stem cells by introducing exogenous Oct3/4, Sox2, Klf4, and c-Myc (OSKM). The reversed cells own the pluripotency, i.e. self-renewal and differentiating into cells of the three germ layers, in nude mice tumours and then able to develop into all types of cells as properties of ES cells and express ES cell marker genes. This work turned the direction of stem cell research in affirming that iPS cells are free from the ethical issues that plague the human cloning and embryonic stem cells. Other following studies show that iPSC can form any kind of differentiated cell, can be derived from anyone.

Currently many scientists could have such a near-limitless supply of iPS cells. During one small-table discussion last year, they mentioned that they have more than one bank of collections of 2000 iPS cell lines. Then he said the first iPS clinic application will be in 2 years in Japan. Logically, these iPS cells have the identical genetics background to the person from whom they are derived. So, once the differentiated cells coaxed from iPS cells, they could be transplanted back into a patient and assumed no rejections. However, the percentage in mixed cell population is so low for iPSC that they are insufficient for therapeutic application and techniques advancements will need to resolve this bottleneck problem.

## The Significance of the Simple but Versatile iPSC Technology

These iPS cells are also awesome disease models. At the cellular level, the iPS cells can essentially become the proxy of 'the patient' with genetic diseases [1]. They may carry the same disease-causing mutations, genetic variants present in patient. Scientists may model the disease more accurately, for instance, which genes go awry with an arrays of details, such as the conditions, the timing, the causes, and progression and then may find ways to put the brake for diseases in their tracks. The procedure is to take some skin cells from a person with disease and then reprogram them back into iPS cells. Afterwards, to coax these stem cells to turn into desired differentiated cells and watch how they work, how they go wrong, and test out hypothesis, or screen potential drugs. Once the iPS cells differentiate to form the cell

***Corresponding author:** Yue Zhang, PhD., Research Center, CHUM, Notre Dame Hospital, 1560 Sherbrooke Street East, Pavillon DeSève, Room Y2625, Montreal, Quebec, Canada, Tel: 1-514-890-8000 ext 23875; E-mail: zhanglee2006@gmail.com

**Received** December 08, 2012; **Accepted** December 10, 2012; **Published** December 13, 2012

**Citation:** Zhang Y (2012) From "Old" Cloning to "Young" Cellular Reprogramming: Nobel Prize 2012 Spotlighted the Stem Cell Work. Clon Transgen 1:e101. doi:10.4172/2168-9849.1000e101

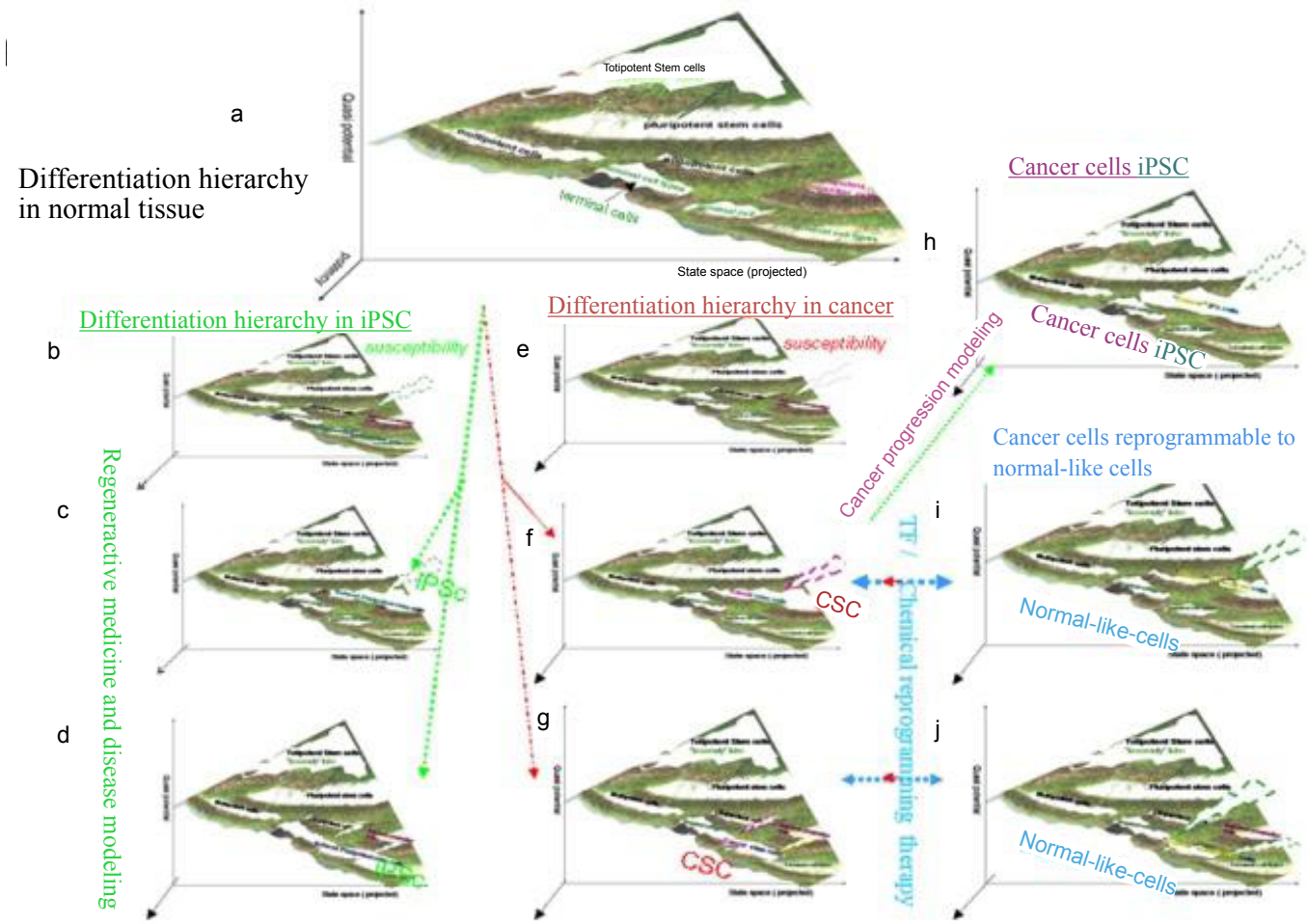


**Figure 1:** "Dams and Lakes" epigenetics landscape and cancer bidirectional reprogramming.
**a: Differentiation hierarchy in normal tissue**
Potency: the potency of cell diversity.
**b-d: Differentiation hierarchy in iPS**
**b**: the intrinsic and external factors shake the constraint of cell attractors.
**c,d**: the over-expression of TFs or CRS deficiency overturns the regulatory constraints and empower the pluripotency to normal cells (c: oligopotent or unipotent cells, d: terminal cells).
**e-g: Differentiation hierarchy in cancer**
**e**: the ageing related or environmental carcinogens, resulting in abnormal expression of TFs or CRS deficiency overturn the constraint and empower the aberrant "pluripotency ' to normal cells (**f**: oligopotent or unipotent cells, **g**: terminal cells).
**h**: Cancer patient induced pluripotent stem cell (iPS). The over-expression of TFs or chemicals, CRS deficiency by epidrug overturns the constraint and empowers the pluripotency to cancer cells.
**i,j: Cancer cell reprogrammable to normal -like cells**
i,j: chemicals or CRS deficiency by epidrug overturn the constraint and empower the aberrant pluripotent cancer stem like cells to normal-like cells**.**
As a derivative of Waddington epigenetics landscape [30], "Dams and Lakes" epigenetics landscape emphasizes the reversibility of cellular reprogramming, particularly we need that:
**First, some definitions:**
**1**) The "cell" is here defined as a unit of (Dam + Lake), but the whole unit will behave similarly along with the downward trend as the "stone" in Waddington epigenetic landscape. Dam: here defined as the locker/gatekeeper of attractors, the regulatory constraints of Gene regulatory network (GRN), the stabilizers of multi-cellular differentiation states. Lake: all other biological characteristics rather than those of Dam in cell, and the "Lake water" represent the majority of proteins, RNAs, and so on.
**2**) The totipotency of cells: the merged "power" of altitude and volume (S) of lake water unit ("Dam and Lake").
**3**) The "lightning" means ageing, the deficient TF expressions, mutations, the tissue disruption, the extra-cellular matrix (ECM) remodelling, the over-expression of TF (e.g. H-RAS, OSKM, etc), the stressors.
**4**) The lightning causes the "heavy rain" and flood, then raises the water level of "lake" or "wave" that may correspond to "higher cellular noise" of gene expression heterogeneity, the driving force of progress of cancer or pluripotency. When the inundated "lakes" merge, then the "dams" could sink under the water. The "lakes" become the bigger and more "powerful" one than their normal counterparts.
**5**) The robustness is that the dam can tolerate a "light rain".
**6**) The "Dam" is made from multiple factors, such as chromatin remodel systems including Mi-2/NuRD, dREAM, the heterochronic genes, ECM genes, the cancer drivers such as p53, PI3KCA, KDM5c, APC, FBXW7, Rb, Ras, VHL, SETD2, mTOR, PTEN, and so on.
**2nd, we could like to hypothesize that**:
**1**) The same tier differentiated cells are easy to reach same potency with direction conversion.
Therefore we could claim that: 1). iPS state might be one "we-want–it" exceptional case among the general cancerous states. 2). Here with a focus on cancer, but it could possibly applied on other complex diseases as well.
Note: In red signifies the malignant reprogramming; in green signifies the benignant reprogramming.

types associated with the illness, the effects of these genetic variants may reveal the underlying mechanisms of its symptoms, progression mechanisms, and look for ways to slow this down or even reverse it [1]. Ideally, it can expose windows of opportunity for treating the condition before symptoms appear, i.e. early prevention. Besides, pharmaceutical industry can join to screen out reagents that restore gene or protein expression to normal levels in its derived cells and then project therapies for disorders with no treatment available. It can also bridge the gaps of diseases with no good animal models, such as heart failure. Drug screening can be improved while established disease iPS cell line models could find out effective targets or minimize the side effects of medications [1]. The personalized medicine may be feasible through benefiting from iPS. Because iPS cells can be derived from any individual of any genetic background, they may eventually tell us the patient-specific disease susceptibility, drug resistance, drug patient-preferential toxicity and so on. Amazingly, iPSC has inspired us to apply direct conversion among differentiated cells and many brilliant findings bursts out [9]. Perhaps one day, we can effectively reverse the diseased biological clocks to a more youthful or healthier state by using a cocktail of chemicals alone [10] or combined approach [11].

## Some Issues of iPSC

Seemingly it is fantastic, but some points we need pay attention to

### The immune reactions

Since iPS cells are genetically identical to their donors' cells, i.e. patient cell, transplants of the cells were assumed not to trigger the body's immune defences, but that view is now being questioned. Last year two interesting studies cause our cautions for iPS clinic trial in that iPS cells triggered immune reactions when they were implanted into mice. In some cases, the cells were completely destroyed by the animals' immune systems [12,13].

### The tumorigenicity

With both self-renewal and differentiating into cells of the three germ layers, the human induced pluripotent stem cells (HiPSCs) are invaluable for regenerative medicine. However, the same properties also make them tumorigenic, and therefore hurdle their clinical application [14]. There are important genetic and epigenetic differences between these ESCs and IPSCs, which seem to influence their tumorigenicity [8,15]. This could allow us to play with epigenetic avenues to deal with such issues [15]. However, a latest brief report seems be encouraging in that chimeric pigs produced from induced pluripotent stem cells demonstrate germline transmission and no evidence of tumor formation in young pigs [16].

### One exceptional but "good" case among cancer-like cells

What is currently encouraging in cancer research is that accumulated evidence is emerging for the feasibility of the bidirectional reprogramming of cancer cells. Some new studies show that the reprogramming of cancer cells generated "stem-like" cells [17-20], and the therapeutic epigenetic reprogramming transformed cancer cells to somehow normal-like cells [21], i.e. improvement of their "immature" differentiation [18,22-24]. Further, we have recently discussed cell attractors-associated cell divisions, dynamics of germline genes' expression and cellular reprogramming within a context of origin of life, evolution and tumorigenesis alongside the evolution and at a system level in the nematode *Caenorhabditis elegans,* fruit fly *Drosophila melanogaster* and mouse "model" organisms. The concept of chromatin remodelling system (CRS)-linked "stem-like" cancer attractors may naturally explain germ-line gene-reactivated carcinogenesis [24-27], and point out the significance of cancer cell education. Broadly to say, we may consider the iPS cell as one exceptional case of the bidirectional reprogramming of cancerous cells (Figure 1). The bidirectional reprogramming and dynamic action of gene regulation in the Mi-2/NuRD complex and its related CRS may provide a resource for investigating carcinogenesis, cancer progress and tumor heterogeneity (Figure 1).

Now it is basically feasible to interfere with the program of germline genes re-activated expression to blunt the growth of the tumors with epidrugs (e.g. DNMTs', HDACs' inhibitors alike) reprogramming. The epigenetic reprogramming along with novel chemical screening iPS cancer cells model as aforementioned will hold a promise for the prognosis and diagnosis of cancer at early stage.

### Elite and stochastic model argument

Dezawa lab reported Multilineage-differentiating stress-enduring (Muse) cells are a primary source of induced pluripotent stem cells in human fibroblasts, which express stem cell-like properties can be reprogrammed [28], thus instead supporting the elite model. The stochastic model has been discussed for a while [29]. For now, we need an open to that the reprogramming process perhaps lies somewhere between the two.

Therefore, now iPS technology, along with a risk but a bit like human "artificial selection", empowers us to look "back" at the "archaic" life forms and look "ahead "at life forms in the future with "cell evolution" in dish. Thus it seems be one addition to the standard natural selection that can make organisms to evolve. In short, both the impact of Gurdon's experiments and iPS enables us to challenge nature and to break boundaries.

### References

1. Rossant J, Mummery C (2012) Nobel 2012 Physiology or medicine: Mature cells can be rejuvenated. Nature 492.
2. Yu J, Vodyanik MA, Smuga-Otto K, Antosiewicz-Bourget J, Frane JL, et al. (2007) Induced pluripotent stem cell lines derived from human somatic cells. Science 318: 1917-1920.
3. Campbell KH, McWhir J, Ritchie WA, Wilmut I (1996) Sheep cloned by nuclear transfer from a cultured cell line. Nature 380: 64-66.
4. Liao L, Li L, Zhao RC (2007) Stem cell research in China. Philos Trans R Soc Lond B Biol Sci 362: 1107-1112.
5. Yue Z, Xinping Z (2002) Progress of fish gene technology research in China. Aquaculture Asia 7: 15-6.
6. Gurdon JB (1962) The developmental capacity of nuclei taken from intestinal epithelium cells of feeding tadpoles. J Embryol Exp Morphol 10: 622-640.
7. Craven L, Tuppen HA, Greggains GD, Harbottle SJ, Murphy JL, et al. (2010) Pronuclear transfer in human embryos to prevent transmission of mitochondrial DNA disease. Nature 465: 82-85.
8. Takahashi K, Yamanaka S (2006) Induction of pluripotent stem cells from mouse embryonic and adult fibroblast cultures by defined factors. Cell 126: 663-676.
9. Efe JA, Hilcove S, Kim J, Zhou H, Ouyang K, et al. (2011) Conversion of mouse fibroblasts into cardiomyocytes using a direct reprogramming strategy. Nat Cell Biol 13: 215-222.
10. Ellis RE (2010) Stem cells: chemically reprogramming cell fates. Nat Chem Biol 6: 84-85.
11. Pereira CF, Lemischka IR, Moore K (2012) Reprogramming cell fates: insights from combinatorial approaches. Ann N Y Acad Sci 1266: 7-17.
12. Okita K, Nagata N, Yamanaka S (2011) Immunogenicity of induced pluripotent stem cells. Circ Res 109: 720-721.

13. Zhao T, Zhang ZN, Rong Z, Xu Y (2011) Immunogenicity of induced pluripotent stem cells. Nature 474: 212-215.

14. Yamashita T, Kawai H, Tian F, Ohta Y, Abe K (2011) Tumorigenic development of induced pluripotent stem cells in ischemic mouse brain. Cell Transplant 20: 883-891.

15. Doege CA, Inoue K, Yamashita T, Rhee DB, Travis S, et al. (2012) Early-stage epigenetic modification during somatic cell reprogramming by Parp1 and Tet2. Nature 488: 652-655.

16. West FD, Uhl EW, Liu Y, Stowe H, Lu Y, et al. (2011) Brief report: chimeric pigs produced from induced pluripotent stem cells demonstrate germline transmission and no evidence of tumor formation in young pigs. Stem Cells 29: 1640-1643.

17. Greathouse KL, Bredfeldt T, Everitt JI, Lin K, Berry T, et al. (2012) Environmental estrogens differentially engage the histone methyltransferase EZH2 to increase risk of uterine tumorigenesis. Mol Cancer Res 10: 546-557.

18. Baylin SB, Jones PA (2011) A decade of exploring the cancer epigenome - biological and translational implications. Nat Rev Cancer 11: 726-734.

19. Lang JY, Shi Y, Chin YE (2012) Reprogramming cancer cells: back to the future. Oncogene.

20. Miyoshi N, Ishii H, Nagai K, Hoshino H, Mimori K, et al. (2010) Defined factors induce reprogramming of gastrointestinal cancer cells. Proc Natl Acad Sci U S A 107: 40-45.

21. Tsai HC, Li H, Van Neste L, Cai Y, Robert C, et al. (2012) Transient low doses of DNA-demethylating agents exert durable antitumor effects on hematological and epithelial tumor cells. Cancer Cell 21: 430-446.

22. Lipkin G (2008) Plasticity of the cancer cell: implications for epigenetic control of melanoma and other malignancies. J Invest Dermatol 128: 2152-2155.

23. Schenk T, Chen WC, Göllner S, Howell L, Jin L, et al. (2012) Inhibition of the LSD1 (KDM1A) demethylase reactivates the all-trans-retinoic acid differentiation pathway in acute myeloid leukemia. Nat Med 18: 605-611.

24. Zhang Y (2012) Ageing and cancer: breaking the “Don't put all eggs in one basket” and natural “self-organisation”, and their potential reprogramming via modulation of Mi-2/NuRD and mTOR kinase. Enzyme Engineering.

25. Xia XM, Yu P, Peng N, Zhang Y, Xue YN, et al. (2011) Tunable release of biomacromolecules from reductive-responsive multilayered hollow microcapsules. J Control Release 152: e101-e103.

26. Zhang Y (2011) Biology of the Mi-2/NuRD Complex in SLAC (Stemness, Longevity/Ageing, and Cancer). Gene Regul Syst Bio 5: 1-26.

27. Zhang Y, Moriguchi H (2011) Chromatin remodeling system, cancer stem-like attractors, and cellular reprogramming. Cell Mol Life Sci 68: 3557-3571.

28. Wakao S, Kitada M, Kuroda Y, Shigemoto T, Matsuse D, et al. (2011) Multilineage-differentiating stress-enduring (Muse) cells are a primary source of induced pluripotent stem cells in human fibroblasts. Proc Natl Acad Sci U S A 108: 9875-9880.

29. Yamanaka S (2009) Elite and stochastic models for induced pluripotent stem cell generation. Nature 460: 49-52.

30. Huang S (2011) On the intrinsic inevitability of cancer: from foetal to fatal attraction. Semin Cancer Biol 21: 183-199.