Available online at www.sciencedirect.com

ScienceDirect

Current Opinion in Cell Biology

Advancing insights into stem cell niche complexities with next-generation technologies

Nicholas Heitman[1,2,4,5], Nivedita Saxena[1,2,4,5] and Michael Rendl[1,2,3,4]

Adult tissue-specific stem cells are essential for homeostatic tissue maintenance and key to regeneration during injury repair or disease. Many critical stem cell functions rely on the presence of well-timed cues from the microenvironment or niche, which includes a diverse range of components, including neuronal, circulating and extracellular matrix inputs as well as an array of neighboring niche cells directly interacting with the stem cells. However, studies of stem cells and their niche have been challenging due to the complexity of adult stem cell functions, their intrinsic controls and the multiple regulatory niche components. Here, we review recent major advances in our understanding of the complex interplay between stem cells and their niche that were enabled by the tremendous technological leaps in single-cell transcriptome analyses, 3D *in vitro* cultures and 4D *in vivo* microscopy of stem cell niches.

Addresses
[1] Black Family Stem Cell Institute, Icahn School of Medicine at Mount Sinai, Atran Building AB7-10C, Box 1020, 1428 Madison Ave, New York, NY 10029, USA
[2] Department of Cell, Developmental and Regenerative Biology, Icahn School of Medicine at Mount Sinai, Atran Building AB7-10C, Box 1020, 1428 Madison Ave, New York, NY 10029, USA
[3] Department of Dermatology, Icahn School of Medicine at Mount Sinai, Box 1047, One Gustave L. Levy Place, New York, NY 10029, USA,
[4] Graduate School of Biomedical Sciences, Icahn School of Medicine at Mount Sinai, Box 1022, One Gustave L. Levy Place, New York, NY 10029, USA

Corresponding author: Rendl, Michael (michael.rendl@mssm.edu)
[5] Both authors contributed equally to this work.

Current Opinion in Cell Biology 2018, **55**:87–95

This review comes from a themed issue on **Differentiation and disease**

Edited by **Katja Röper** and **Xosé R Bustelo**

https://doi.org/10.1016/j.ceb.2018.06.012

Introduction

Each day, an adult human generates billions of new cells to replace those that are lost naturally or by damage. At the top of the production hierarchies are adult stem cells (SCs), defined by their abilities for long-term self-renewal and multipotent differentiation into several different lineage-restricted cell types. The first definitive proof of the existence of adult SCs came from work by Till, McCulloch and others in the 1960s demonstrating the existence of a hematopoietic SC (HSC) pool responsible for maintaining the entire blood-lineage throughout life [1–3]. Since then, multiple adult SCs have been discovered in several tissues and organs, such as the intestine [4,5], brain [6,7], mammary gland [8,9], and skin [10] including hair follicles [11–13].

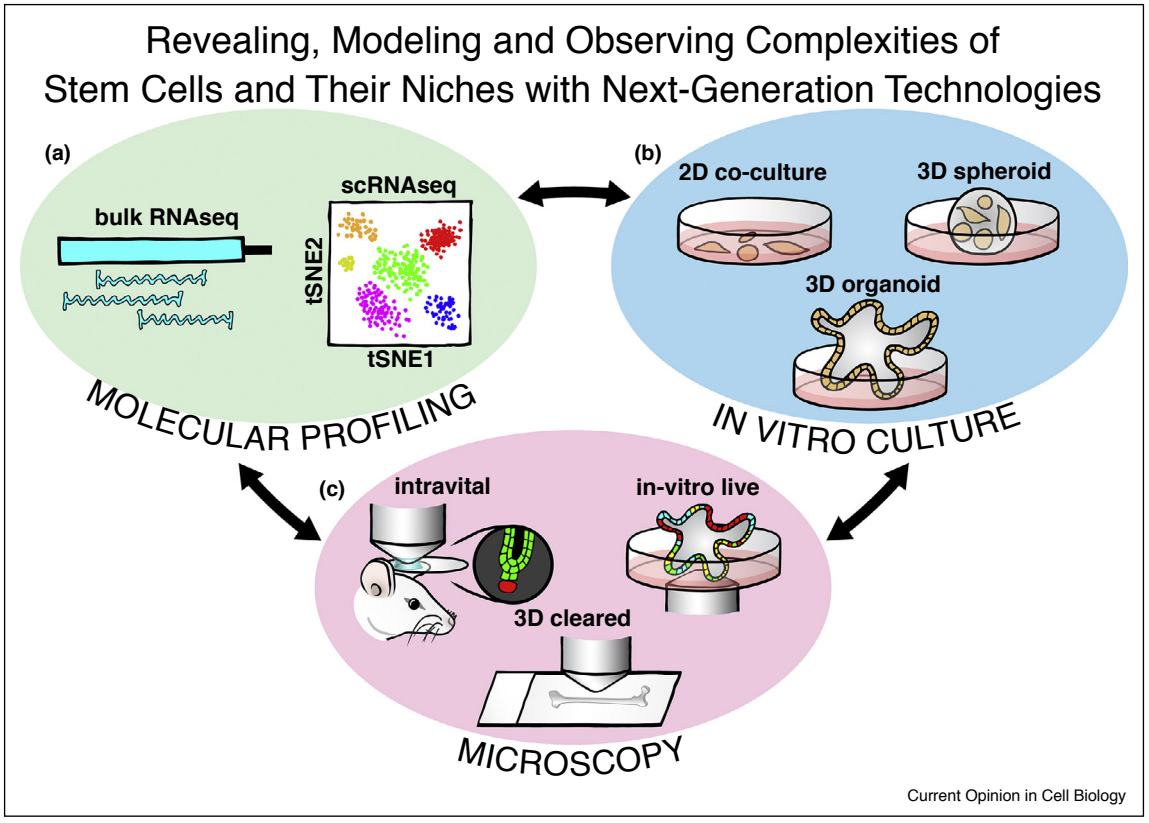
Although many of the special characteristics of SCs are intrinsic, no completely autonomous adult SC has been discovered. Rather, all known adult SCs rely to a large extent for proper function on external signals from its surroundings, termed the SC niche [14–16]. The niche communicates vital information regarding the regenerative needs of the tissue, the importance of which is exemplified by the detrimental effects of deviations from the crucial loss/production balance, as defects of SCs and their niches have been implicated in multiple human disorders and diseases [17].

The HSC sits atop a hierarchy of fate-committed multipotent progenitors (MPP) and terminally differentiated cells of the entire blood-lineage [18–20]. Self-renewal of HSCs and lineage-committed progenitors and their differentiation towards diverse blood lineages are regulated by multiple niche inputs from non-hematopoietic cell types, such as osteoblasts [21,22], peri-sinusoidal [23,24] and peri-arteriolar stromal cells [25], and endothelial cells [23,26], as well as from hematopoietic lineages, such as macrophages [27–29] and megakaryocytes [30].

Hair follicle SCs (HFSC) and downstream progenitors give rise to seven cell lineages that make up the hair shaft and its supporting channel during hair growth, a process that is interrupted by a naturally occurring hair cycle of cyclical bouts of follicle destruction, a resting phase of relative quiescence, and re-growth [11–13,31]. Distinct niche signals controlling the balance of SC rest and activation are thought to emanate from essential niche components that include specialized mesenchymal dermal papilla cells [32–35], direct SC progeny of multipotent progenitors [36,37••] and neighboring nerves [38], as well as longer-range inputs from fibroblasts deep in the dermis [39], cells of the dermal adipocyte-lineage [40], and immune cells [41–43].

Intestinal SCs (ISC) residing in the intestinal crypt base constantly replenish the villus epithelium of rapidly

Figure 1



Next-generation technologies, often used in combination or complementation, advance insights into SC niche complexities. **(a)** Molecular profiling by population-based RNA sequencing and by single-cell RNA sequencing reveal unprecedented level of gene expression complexities and cell heterogeneity in SCs and their niches. **(b)** *In vitro* cultures with 2D engineered matrices and co-culture, 3D spheroid aggregates, and structured 3D organoids enable modeling of SC interactions with their niches. **(c)** 3D multicolor light microscopy of 3D cleared tissues and 4D *intravital* and *in vitro* live imaging allow observation of SC niche complexities *in vivo* and in real time.

turned-over enterocytes, goblet and enteroendocrine cells, and other SC progeny that are lost by conveyor belt-like upward displacement [4,5,44] The intestinal regulatory niche contains pericryptal mesenchymal fibroblasts [45,46], myofibroblasts and smooth muscle cells [47,48], as well as gut lumen microbiota [49,50]. Paneth cells, the only differentiated SC progeny that migrate to the crypt base, also provide regulatory niche signals to ISCs [51–53].

In this review we highlight recent major discoveries into the SC regulation by the niche that were made possible by groundbreaking new technological innovations of single cell-level profiling, complex cell and organoid cultures and major advances in light microscopy (Figure 1). As we are not able to cover the entire extensive body of new knowledge gained in the SC field within the past few years, we refer to excellent recent reviews containing comprehensive updates in several SC niche systems [54–58].

Revealing complexity: single-cell profiling of organs

Precise regulation of gene expression in SCs and their niche is paramount for executing the molecular programs of SC quiescence, self-renewal and lineage differentiation. Specific sets of expressed genes and epigenetic configurations underlie functional distinctions between different cell types within complex tissues, including SCs and the cellular niche components. Since large-scale transcriptome analysis became technically feasible with the establishment of microarrays in *Arabidopsis* [59], it has been used with great impact as a window into SC and niche-specific properties and as a basis for discovering targets for functional studies in multiple SC niche systems [11,12,60–62]. Since then, technologies committed to monitoring the transcriptome of cells, as surrogate for protein expression, have flourished. RNA-sequencing (RNAseq) was established in rapid succession in *Arabidopsis* [63], yeast [64] and mammalian cells [65] that surveys mRNA content in a manner that is relatively