by single-cell sequencing (Olsson et al., 2016) have functional roles in either patch or cluster formation. Perhaps more importantly, these unique populations that comprise pGMPs and cGMPs can be targeted in leukemia or activated in myelosuppressive disorders by the implementation of novel therapeutic approaches. Collectively, this study advances our understanding of myeloid progenitor biology and of the BM microenvironment, with important therapeutic implications in hematological disease.

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Synthetic Embryos: Windows into Mammalian Development

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Mammalian embryonic stem cells (ESCs) can self-organize in vitro, but whether they can recreate early embryonic morphogenesis is unclear. Harrison et al. recently demonstrate in *Science* that 3D co-cultures of mouse ESCs and trophoblast stem cells self-organize into embryo-like structures that recreate many features of early mouse development.

The early mammalian embryo possesses a remarkable ability to self-organize. In contrast to other well-studied models such as the fly or frog, in which a maternal deposition of mRNA or protein is the initial symmetry-breaking event, mammalian embryos appear to establish the body axes independently of any maternal cues. Mammals begin life by creating a ball of nearly identical cells that then proliferate and undergo a series of decisions, forming two extraembryonic tissues, the trophectoderm and extraembryonic endoderm, and the pluripotent cells of the epiblast that will give rise to the embryo itself. During gastrulation, the epiblast subdivides into the three germ layers under the influence of signals issuing from the extraembryonic cells. The fact that these events occur in a self-organized fashion means that it may be possible to recreate them in vitro beginning from embryonic stem cells (ESCs), and recent studies show that both three-dimensional aggregates of mouse ESCs and two-dimensional colonies of human ESCs grown in defined geometries have the ability to organize all three embryonic germ layers (van den Brink et al., 2014; Warmflash et al., 2014). Nonetheless, these systems do not clearly establish body axes, and to the extent that they undergo morphogenesis, their overall form is quite different from that of embryos. These shortcomings may be due to the absence of interactions between the extraembryonic and embryonic tissues, which are required to properly establish the body axes of the embryo in vivo (Arnold and Robertson, 2009).

In a recent article published in *Science*, Magdalena Zernicka-Goetz and colleagues report an important step forward in recreating embryo-like structures in vitro (Harrison et al., 2017). Using Matrigel as a scaffold, they combine mouse ESCs and trophoblast stem cells (TSCs) in three-dimensional culture, and they find that when these populations come together, they self-organize into a structure resembling the mouse embryo with segregated populations of ESCs and TSCs (called an ETS embryo). Amazingly, these ETS embryos undergo much of the morphogenesis of the mouse embryo at the egg cylinder stage. Similar to their in vivo counterparts, a cavity begins to form in the embryonic portion of ETS embryos, followed by a cavity in the extraembryonic portion, and these two cavities then fuse. While the ESC cavity appears to form autonomously, Nodal signaling emanating from the ESCs is required to induce cavity formation in the TSCs. ETS embryos also initiate important features of later development including asymmetric expression of primitive streak markers and germ cell formation on the interface between ESCs and TSCs. The

ETS embryo system thus represents an exciting opportunity to study the complex morphogenesis of the egg cylinder stage and the initiation of gastrulation processes in vitro.

These results reveal a number of surprising properties of early mammalian development. The ability to recreate much of the morphogenesis of the epiblast-staged embryo simply by juxtaposing ESC and TSC populations suggests that the precise organization of the blastocyst stage is dispensable for later development. Second, ETS embryos show remarkable consistency in size, and they appear to initiate gastrulation-like processes when their size is nearly the same as that of gastrulation-staged mouse embryos. This may point to mechanisms of size-sensing that trigger further development when the ETS embryo reaches an appropriate size. As ESC aggregates do not show the same size consistency, it is interesting to speculate that size control may depend on interactions between ESCs and TSCs. Finally, these ETS embryos show two clear axes: the proximal-distal one, which is defined by the position of the TSCs relative to the ESC compartment, and the anterior-posterior axis, which self-organizes and is defined by the markers of the primitive streak and germ cells. It is surprising that cells are able to position a primitive streak asymmetrically in the absence of the extraembryonic endoderm, given the requirement for secreted inhibitors from the anterior visceral endoderm (AVE) in this process in vivo (Perea-Gomez et al., 2002). It seems likely that the primitive streak region, once induced, can inhibit formation of the same structure on the other side of ETS embryos; however, it remains an open question why the same mechanism doesn't break symmetry in embryos lacking the secreted inhibitors, in embryos in which the AVE fails to form (Nowotschin et al., 2013), or in micropatterned human ESC cultures (Warmflash et al., 2014).

It will be exciting to follow up on this initial characterization to more fully understand the development of ETS embryos and its relationship to normal embryogenesis. One key issue is that of timing and whether ETS embryos can be precisely staged with a correspondence to natural embryo development. In this study, ETS embryos from a relatively broad temporal window of 72–96 hr in culture are compared to epiblast-staged embryos (E5.5–5.75); however, expression of Stella, a marker of germ cells that is only detected in vivo starting at E7.5 (Payer et al., 2006), is also detected by 96 hr. It will also be interesting to probe both earlier and later events. Earlier stages of ETS development clearly do not replicate natural development as, in contrast to natural embryos, the ESC and TSC compartments are separated from the start, but they could still shed light on the mechanisms of self-organization that generate the epiblast-like configurations. How is it that ESCs and TSCs are programmed to recreate their embryonic architecture even from relatively unstructured initial conditions? It is unclear from the current study how far into development ETS embryos can proceed. Do three germ layers form, and do cells migrate out of the primitive streak in the anterior direction? The remarkable emergence of both the primitive streak and germ cells appears to be somewhat less organized and to involve fewer cells than their in vivo counterparts. It is to be expected that, at some point, ETS and natural embryos will correspond less closely, but much can be learned by identifying the points of divergence. These points also represent exciting opportunities to learn about development by replicating it in vitro. If the current culture conditions do not permit certain features to emerge normally, identifying improvements can shed light on the minimal information from outside the embryo proper that is required.

In this regard, the system developed here can also be used to dissect the requirements for communication between the embryonic and extraembryonic compartments. For example, the authors show that the induction of germ cells from ESCs requires BMP signaling and, based on previous work in vivo (Arnold and Robertson, 2009), BMP signaling is likely to be involved in initiating primitive streak formation as well. Can the TSC population in these studies be replaced by a localized source of BMP ligands? If not, what other molecular factors are required?

Finally, it will be exciting to see if a similar protocol can be performed with human ESCs. Doing so may require overcoming technical challenges, as human TSC lines are not available and there are significant differences between human and mouse in the architecture and relative positions of the epiblast and trophoblast populations. Nonetheless, this research is crucial, as even with recent advances in culturing human embryos (Deglincerti et al., 2016; Shahbazi et al., 2016), ethical and practical limitations remain, and synthetic embryos may be the best window into our own development.

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