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How can you tell that stem cells are good at math? You can watch them multiply and divide!

Blood production is an essential process in all vertebrate animals that needs to occur throughout an organism's lifespan; red blood cells need to carry oxygen to distant tissues, platelets are required for stopping bleeding after injuries, and a multitude of other blood cells continually protect the body from foreign invaders. This process is under tight molecular control, as too little or too much blood generation is detrimental to the organism. The key cells that give rise to blood are known as hematopoietic stem cells (HSCs); they arise during embryonic development and are responsible for this constant blood production. HSCs have the unique feature of generating more of themselves (so that the system is never depleted) and also generating other blood progenitors that eventually give rise to mature blood cells.

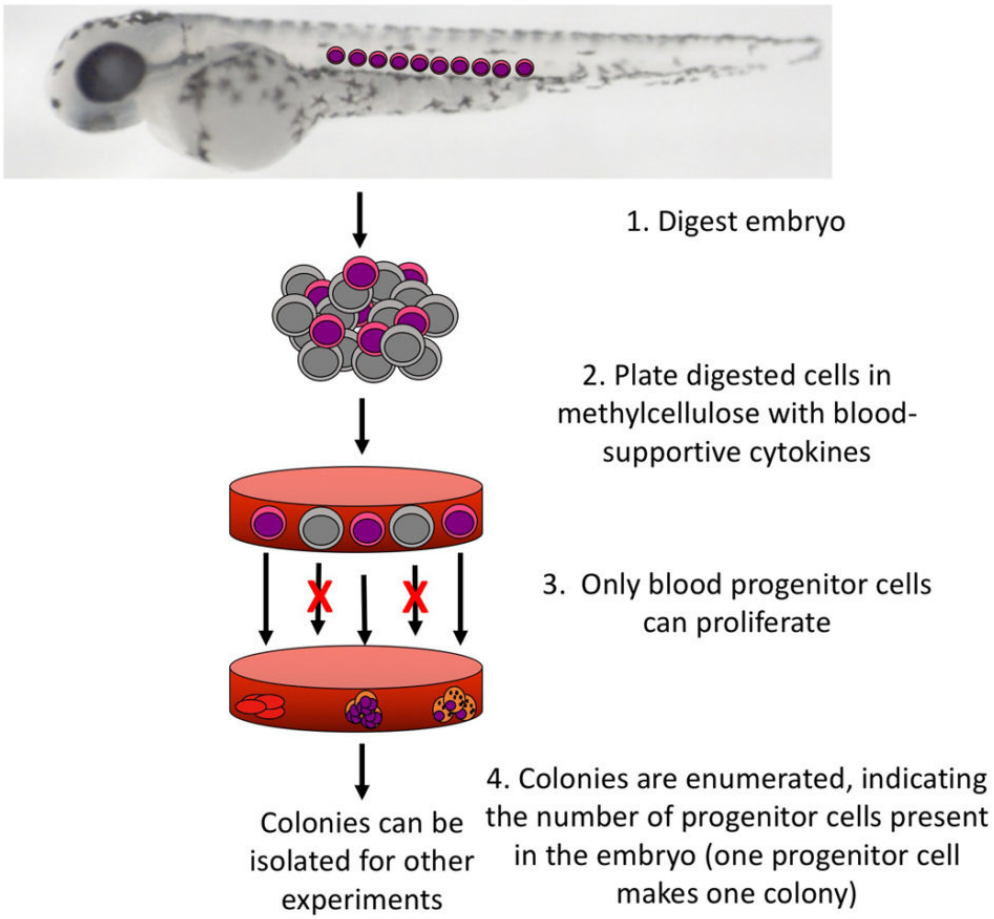

Fig. 1. Counting blood progenitors in the developing embryo. Embryos are enzymatically digested (1), and all cells are plated in methylcellulose, a viscous media that only allows blood cells to grow (2,3). After 7 days, colonies can be counted, which indicates the numbers of blood progenitors that were present in the embryo originally (4). These colonies can be isolated from the methylcellulose for further study, if desired.

HSCs and their downstream progenitors are important therapeutically, as they are the key to repopulating the blood system; defects in these cells are responsible for a lack of blood cells causing diseases like anemia or neutropenia and they are also responsible for leukemia, a family of serious blood cancers. Due to this fact, a large focus in the field is to determine molecular signals that could be used to either make new progenitor cells or isolate, repair, and expand a patient's existing progenitor cells. To do this, scientists turn to animal systems that are easily to study and manipulate, but still similar enough to humans to have clinical relevance. One popular animal system in use is the zebrafish (*Danio rerio*). Zebrafish develop outside of the body and are transparent, so scientists can observe blood development under a microscope as it happens in real time. Every time a zebrafish lays eggs it generates hundreds of offspring, which can be collected, manipulated, and studied. Their blood system is similar enough to humans so that findings in zebrafish are relatable to human health. For these reasons, scientists use zebrafish to perform large-scale experimental screens with the hope of finding compounds and genes that affect the number of blood progenitors. In fact, many compounds have been identified in this manner that are being tested in human clinical trials.

To observe progenitors is difficult, requires expensive equipment, and is time consuming. So, we developed an assay that is quicker, easier, and cheaper, with the hope that more scientists will be able to test their favorite drug compound (or modify their favorite gene) to see its effect on blood progenitors. Basically, all you do is digest the embryo, give it the right growth conditions, and wait a week for the progenitors grow and divide (Fig. 1). After that time, you can count the cells to see how many progenitors were present in the fish; one colony derives from a single progenitor cell. In this way, you can easily treat an embryo with a drug or modify a particular gene before they are subjected to the assay to see if that change will affect the number of progenitor cells present.

Overall, this manuscript describes (and the associated video visually shows) scientists how to count blood progenitors. We hope that this simple, inexpensive assay will allow further discoveries that will help develop treatments to help humans with blood disorders.

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Publication

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