

[0009] Thus, what is needed are methods, compositions and apparatuses for generating patient derived tissue models (e.g., tumor models and/or non-tumor tissue models) from resection or biopsies. In particular, it would be useful to provide methods and apparatuses that may enable a large number of patient-derived tissue models having predictable and clinically relevant properties from a single biopsy, such as an 18-gauge core biopsy, which could be completed within, e.g., 7-10 days after obtaining a biopsy. This would permit robust and reliable testing and minimize delays in guiding patient-specific therapies. Furthermore, it will also be useful to generate patient derived models that can expand quickly in a highly parallel manner, generating units with smaller and more uniform sizes, better controllability for cell number per unit, and better diffusibility (e.g., via increase surface to volume ratio), for high-throughput screen applications.

SUMMARY

[0009a] In one aspect, the present disclosure refers to a method, the method comprising:
dissociating a tissue sample obtained from a patient tumor biopsy or a resected patient tumor sample, thereby forming a dissociated tissue sample, wherein the dissociated tissue sample comprises tumor cells and one or more of mesenchymal cells, endothelial cells, and immune cells resident in the patient tumor biopsy or resected patient tumor sample;
combining the dissociated tissue sample and a fluid matrix material to form an unpolymerized mixture;
forming a plurality of droplets of the unpolymerized mixture by driving a continuous stream of the unpolymerized mixture traveling at a first flow rate across one or more streams of an immiscible fluid traveling at a second flow rate, wherein the immiscible fluid is immiscible with the unpolymerized mixture, and wherein the first flow rate and the second flow rate are different; and
polymerizing the plurality of droplets to form a plurality of spherical droplets each having a diameter of between 50 and 500 μm with between 1 and 200 dissociated cells distributed therein, wherein the spherical droplets comprise three-dimensional cellular structures formed from the tumor cells and the one or more of mesenchymal cells, endothelial cells, fibroblasts, and immune cells resident in the patient tumor biopsy or resected patient tumor sample, wherein the three-dimensional cellular structures replicate a three-dimensional microenvironment of the patient tumor biopsy or resected patient tumor sample from which the spherical droplets were derived.

[0010] Described herein are Patient-Derived Micro-Organospheres (PMOSs), apparatuses and methods of making PMOSs, and apparatuses and methods of using PMOSs. Also described herein are methods and systems for screening a patient using these Patient-Derived Micro-Organospheres, including personalized therapy methods.

CLAIMS

What is claimed is:

1. A method, the method comprising:
 - dissociating a tissue sample obtained from a patient tumor biopsy or a resected patient tumor sample, thereby forming a dissociated tissue sample, wherein the dissociated tissue sample comprises tumor cells and one or more of mesenchymal cells, endothelial cells, and immune cells resident in the patient tumor biopsy or resected patient tumor sample;
 - combining the dissociated tissue sample and a fluid matrix material to form an unpolymerized mixture;
 - forming a plurality of droplets of the unpolymerized mixture by driving a continuous stream of the unpolymerized mixture traveling at a first flow rate across one or more streams of an immiscible fluid traveling at a second flow rate, wherein the immiscible fluid is immiscible with the unpolymerized mixture, and wherein the first flow rate and the second flow rate are different; and
 - polymerizing the plurality of droplets to form a plurality of spherical droplets each having a diameter of between 50 and 500 μm with between 1 and 200 dissociated cells distributed therein, wherein the spherical droplets comprise three-dimensional cellular structures formed from the tumor cells and the one or more of mesenchymal cells, endothelial cells, fibroblasts, and immune cells resident in the patient tumor biopsy or resected patient tumor sample, wherein the three-dimensional cellular structures replicate a three-dimensional microenvironment of the patient tumor biopsy or resected patient tumor sample from which the spherical droplets were derived.
2. The method of claim 1, further comprising modifying the cells within the dissociated tissue sample prior to forming the plurality of droplets.
3. The method of claim 1, wherein forming the plurality of droplets comprises forming a plurality of droplets of the unpolymerized mixture of uniform size with less than 25% variation in size.
4. The method of claim 1, wherein the tissue sample comprises cells that are not stem cells.
5. The method of claim 1, wherein the tissue sample comprises a biopsy sample from a metastatic tumor.

6. The method of claim 1, wherein the patient tumor biopsy or resected patient tumor sample comprises both cancer cells and stroma cells.
7. The method of claim 1, wherein combining the dissociated tissue sample and the fluid matrix material comprises disbursing the dissociated tissue sample within the fluid matrix material to a density of less than 5×10^6 cells/ml.
8. The method of claim 1, wherein forming the plurality of droplets comprises forming the plurality of droplets from a continuous stream of the unpolymerized mixture.
9. The method of claim 8, wherein forming the plurality of droplets comprises applying one or more convergent streams of the immiscible fluid to the stream of unpolymerized mixture.
10. The method of claim 1, further comprising removing the immiscible fluid from the plurality of spherical droplets.
11. The method of claim 1, comprising combining the dissociated tissue sample with the fluid matrix material within six hours of removing the tumor biopsy or resected tumor sample from the patient.
12. The method of claim 1, wherein the polymerizing comprises raising the temperature of the plurality of droplets to a temperature greater than 30 degrees C.
13. The method of claim 1, wherein combining the dissociated tissue sample and the fluid matrix material comprises combining the dissociated tissue sample with a basement membrane matrix.
14. The method of claim 1, further comprising screening the plurality of spherical droplets with a plurality of drug compositions within 14 days of acquiring the patient tumor biopsy or resected patient tumor sample from the patient.
15. The method of claim 1, wherein the culturing comprises culturing the plurality of spherical droplets for between 2-14 days to form the three-dimensional cellular structures clusters that replicate the three-dimensional microenvironment of the patient tumor biopsy or resected patient tumor sample.
16. The method of claim 1, further comprising monitoring the flow rate of the unpolymerized mixture to maintain a continuous stream of the unpolymerized mixture.

17. The method of claim 1, further comprising pressurizing the unpolymerized material.