**Exploring Patterns in Cell Shape and Structure**

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# Student Learning Outcomes

1. **Define** the purpose of the *endoplasmic reticulum* and *high-throughput microscopy*
2. **Compare** cell structures using the Allen 3D Cell Viewer
3. **Explain** in your own words the process and applications of the Allen Institute tools used to create a label-free prediction method for determining the location of 3D structures inside the cell, directly from transmitted light images
4. **Design** a future experiment to build on your findings

# **Anticipated** Time

1. Part I: ~ 15 minutes
2. Part II: ~ 20 minutes
3. Part III: ~ 20 minutes
4. Part IV: ~ 30 minutes

# **Part I. What has happened to these cells?**

Dr. G and his summer students want to discover a drug that could be used as an antibiotic against *Delftia acidovorans*, an opportunistic bacterial pathogen that has been found in drains and faucets. They used high-throughput drug screening to test a library of cancer drugs from the National Cancer Institute (NCI) and were excited to find a promising hit, a compound with potentially useful drug properties. This compound is highly active at low doses against *Delftia acidovorans* and not cytotoxic when tested on mammalian cells. However, Dr. G’s students were learning from a friend in another lab how to use a stain to label the endoplasmic reticulum (ER) in live Chinese Hamster Ovary (CHO) cells and noticed that cells treated with low doses of their promising compound have *wildly* variable ER sizes. They tell Dr. G., and together they search for information about ER structure. Dr. G. is a microbiologist and along with his students has limited cell biology experience.

## **Questions**

1. Draw a eukaryotic cell and identify the endoplasmic reticulum (ER). How is this structure different from a prokaryotic cell?
2. Explain to the average adult the function of the ER.

### Find three or four peers and compare your drawings and answers.  Discuss your reasoning.

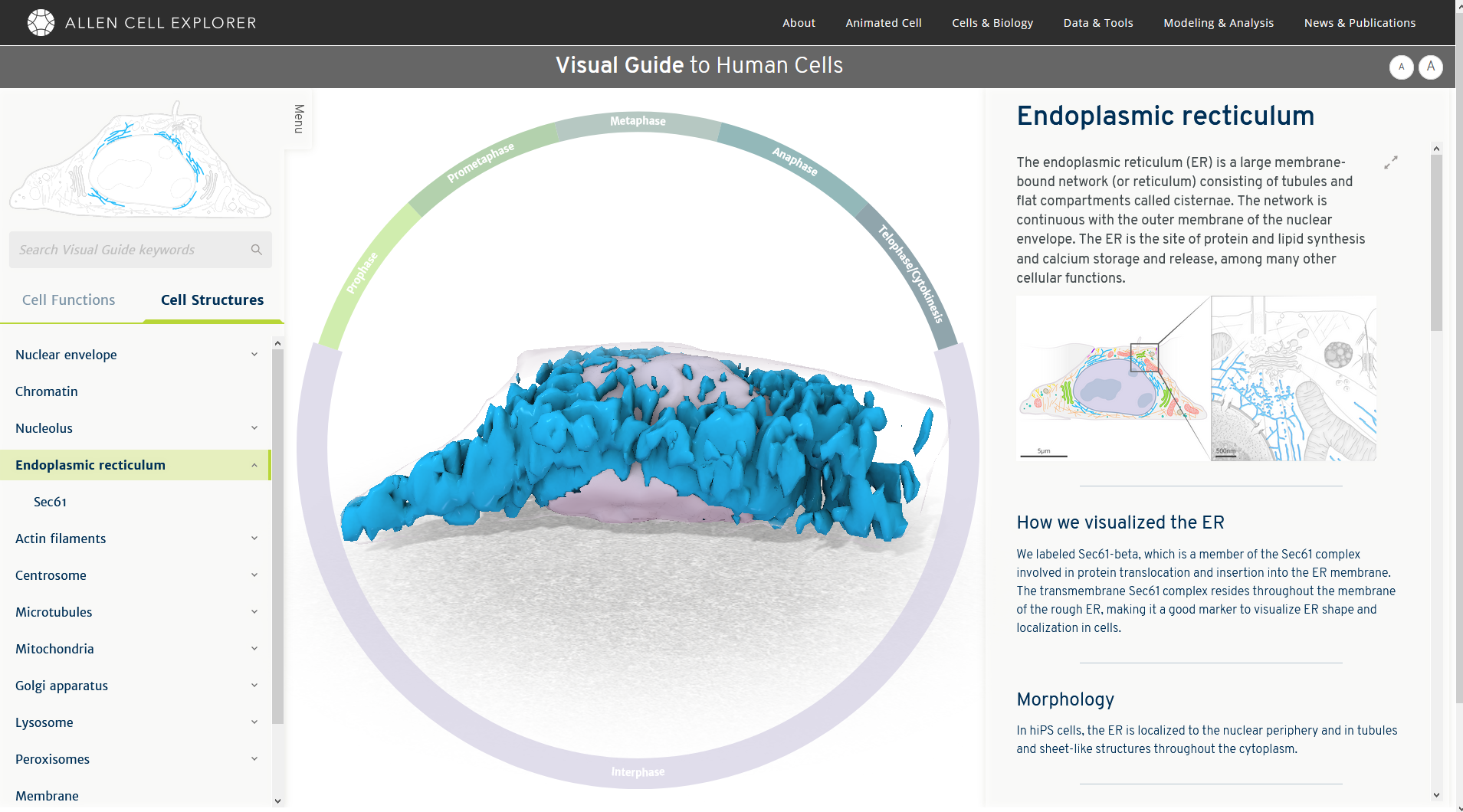
### **STOP HERE!**

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# **Part II. This image does *not* look like the textbook image!**

Dr. G. and his students visit the *Allen Cell Explorer* [*Visual Guide to Human Cells*.](https://www.allencell.org/visual-guide-to-human-cells.html) This resource, along with other open data we will use in this lesson, has been produced by the [Allen Institute for Cell Science](http://cellscience.alleninstitute.org).

They read about the Endoplasmic Reticulum (ER) and how researchers visualized the ER by labeling Sec61-beta proteins as discussed below.

[](https://www.allencell.org/visual-guide-to-human-cells.html)

They then decide to view cells using the [***Cell Feature Explorer***](https://cfe.allencell.org/) and visualize cells with the **Sec61-beta** protein tagged as shown next. Watch this [short video](https://www.youtube.com/watch?v=UKXtEddLzjg) to learn more about the 3D Cell Viewer. These features are all found in the Cell Feature Explorer, along with some additional tools for quantitative analysis of the cells. We’ll be concentrating on the cell images, but you may find additional insights using the graphing panel.

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## **Your turn!**

Help Dr. G. and his students by visiting the Allen Cell Explorer resources. With the information you find, answer the questions below.

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## **Questions**

1. Using the [*Visual Guide to Human Cells*](https://www.allencell.org/visual-guide-to-human-cells.html), look at the shape of the ER during the different phases of the cell cycle (prophase, prometaphase, metaphase, anaphase, and telophase/cytokinesis). You can click on the *name* of the phase, and the visuals will change. *What changes do you notice? (in 2-3 sentences)*
2. You notice under the Morphology section the following text: “In hiPS cells, the ER is localized to the nuclear periphery and in tubules and sheet-like structures throughout the cytoplasm”. Dr. G’s students saw staining using the ER-specific stain that *varied* in shape both within and between different cells. *What explanation do you have for this? (2-3 sentences)*
3. You then navigate to the [*Cell Feature Explorer*](https://cfe.allencell.org), select Sec61-beta under Protein Tag from the menu on the left, and scroll down to view cell images. The number of images is indicated by the number to the right of the protein name. You click on a couple of cell images, keeping in mind that the yellow signal corresponds to a fluorescent tag for the ER and the membrane and DNA are also stained. *Do you notice any patterns? (2-3 sentences)*.

### **STOP!** Find two or three peers and compare your findings. Explain your reasoning.

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# **Part III. Different looking cells? Let’s get quantitative.**

You have been looking at cells for some time now and notice a wide variety of patterns for the Sec61-beta tagged cells. Furthermore, *it seems all cells look different!* This wasn’t what you are used to from your Cell Biology class in college... You want to quantify this as best you can.

In the Cell Feature Explorer, scroll back up to the graph to start measuring the cellular and nuclear volumes of the ER (tagged using the Sec61 protein tag).

## **Questions**

1. Measuring cellular and nuclear volumes. Select three cells and compare the ER volumes. Record the values.

|  |  |  |
| --- | --- | --- |
|  | **Cellular volume** | **Nuclear volume** |
| **Cell 1** |  |  |
| **Cell 2** |  |  |
| **Cell 3** |  |  |

1. Compare your measurements with those obtained by the students next to you.

You want to learn more! You find: [The Integrated Mitotic Stem Cell](https://imsc.allencell.org/).

And carefully scroll down and read… You use the 3D Cell Viewer embedded in the Integrated Mitotic Stem Cell page to visualize the ER and other structures superimposed in space and time.

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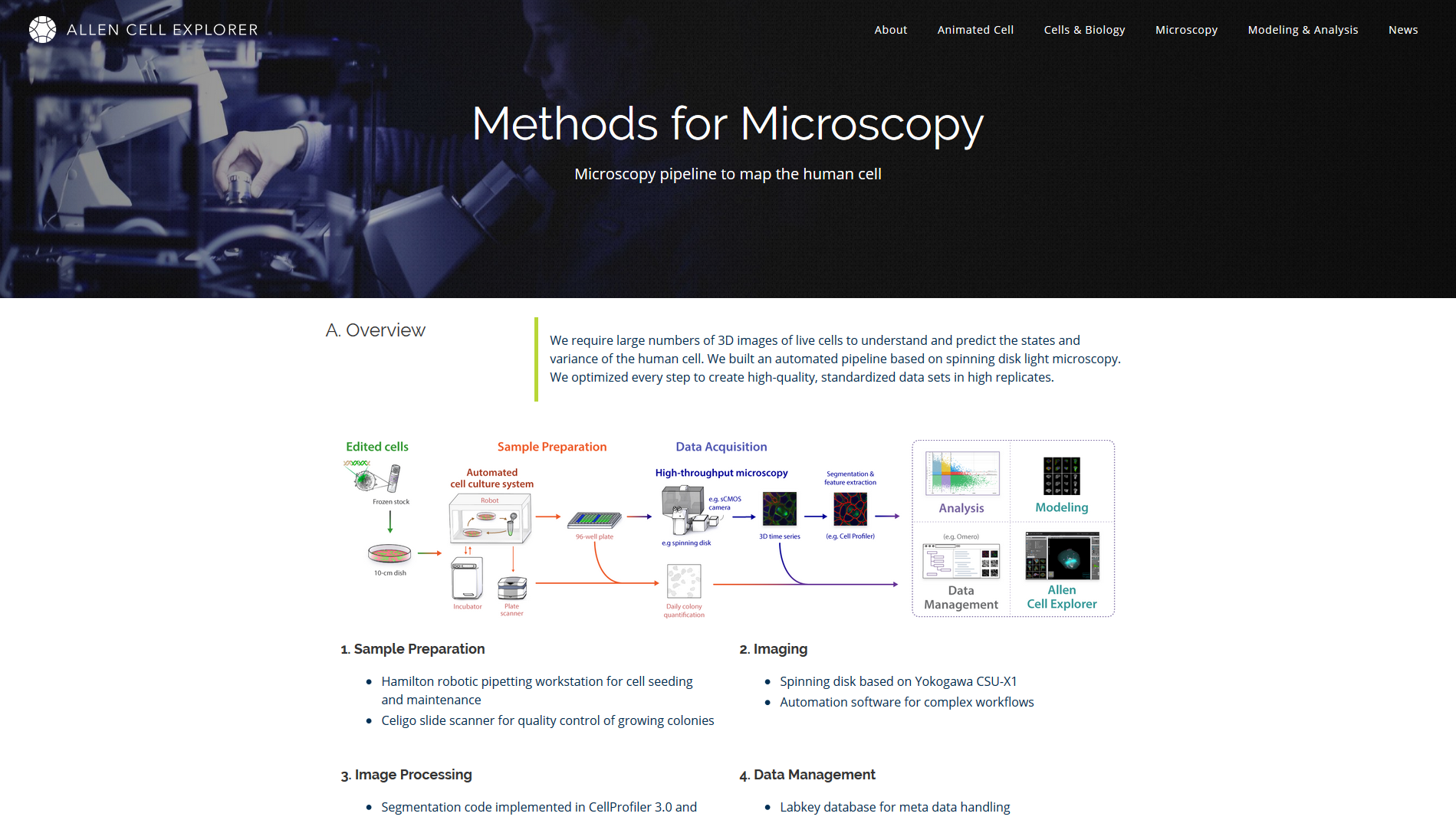
## **Questions**

1. Do these images help you explain the variability in ER volumes you recorded previously?
2. What do you find intriguing?

You start to wonder... *How are they visualizing the ER and other organelles?* You navigate to the [Methods for Microscopy page](https://www.allencell.org/methods-for-microscopy.html) to learn more about the process used by the Allen Institute and begin to wonder...

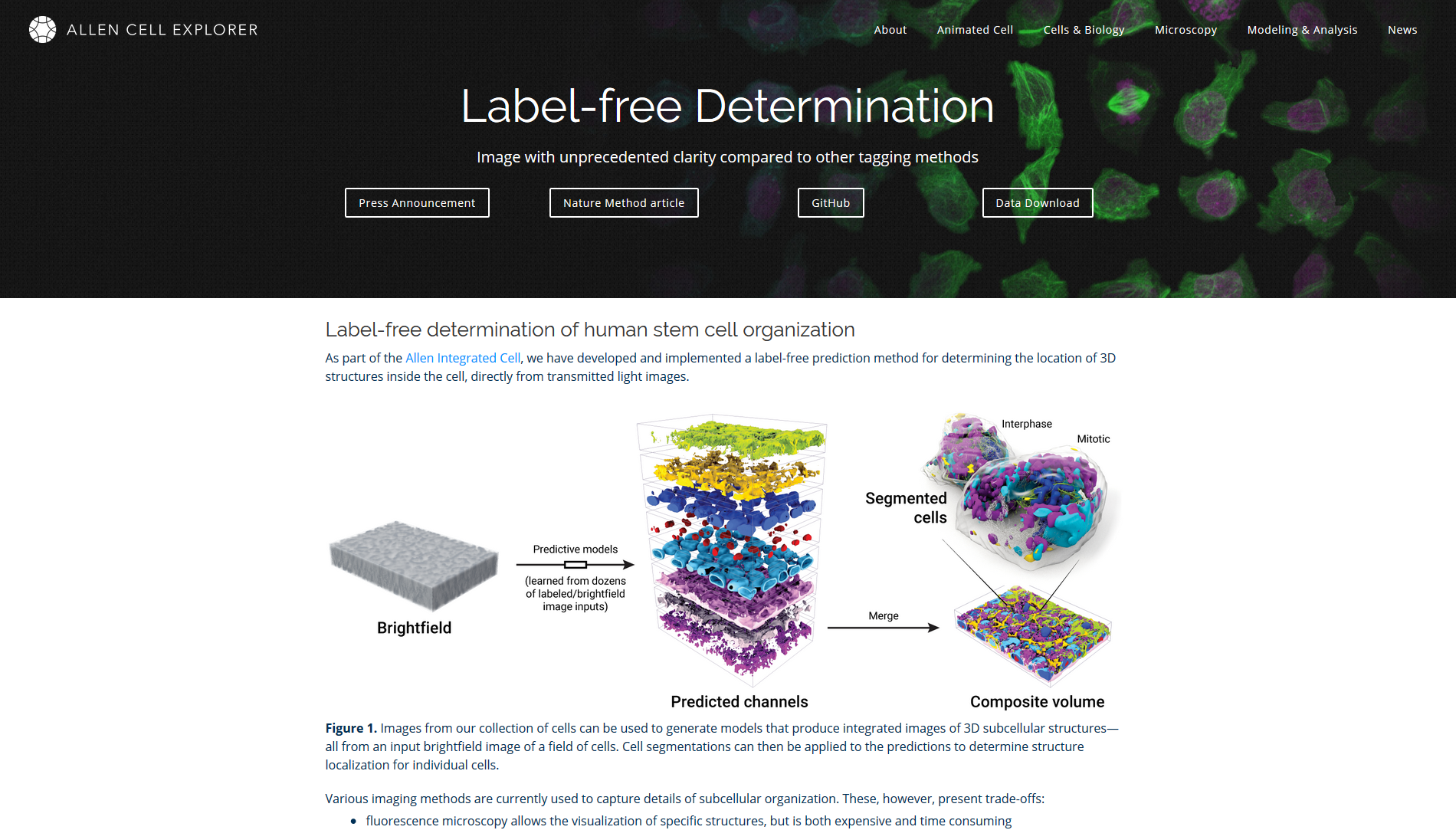
## **Questions**

1. Read the [*Methods for Microscopy* page](https://www.allencell.org/methods-for-microscopy.html). Individually, summarize the methods used in no more than **five** sentences.



You also learn that with this high-throughput microscopy system and the cell lines developed for imaging cell structures, the Allen Institute developed a label-free prediction system.

1. The microscopy methods help a little with your understanding of the imaging process, but you are still curious about the label-free prediction method used to determine the location of structures. You visit the [Label-free Determination](https://www.allencell.org/label-free-determination.html) page to learn more.



Explain *in your own words* the process the Allen Institute used to create a **label**-**free** **prediction** method for determining the location of 3D structures inside the cell, directly from transmitted light. Highlight the main steps involved in the process and how machine learning was used.

### **STOP!** Find two or three peers and compare your findings.

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# **Part IV. Does this make sense, Dr. G?**

You have learned a lot from helping Dr. G’s students and visiting the Allen Cell Explorer. However, you still have to report back to Dr. G.

## **Questions**

1. Think about the high-throughput microscopy approach used by the Allen Institute and the tools developed to measure cell structures. What did you learn from the tools and site?
2. Think about the cells you viewed, what you learned about the ER, and the *Delftia* experiment. *Do you think this high-content imaging approach will help Dr. G’s students? Why or why not? Think about the knowledge gained from this approach and the limitations of the dataset.*
3. If you could work with researchers at the Allen Institute, *what would you do next? Design an* ***experiment*** *to build on your findings and help Dr. G.* Use external resources if necessary.

# **Reflection**

* What was the most **memorable** concept or skill you learned from this case study?
* What are you left **wondering** about [“Nothing” is an unacceptable answer]?How do you start the process of learning the answer?

# **Teaching Notes**

The end of part IV may also be a good moment to talk about the new [Allen Cell Segmenter](https://www.allencell.org/segmenter.html) and challenges of pulling one structure out from microscopy images.

The Allen Institute education outreach page [here](https://alleninstitute.org/about/education-outreach/) includes free posters for classrooms and additional curricula for neuroscience and cell biology. The Allen Institute for Cell Science education resources page [here](https://www.allencell.org/educational-resources.html) includes a video of Carlos presenting on this curriculum and links to resources like the Visual Guide for Human cells.

# Acknowledgements

We appreciate the resources the Allen Institute made available to the public. This case study is part of other cases created as part of the [**NSF HITS RCN**](http://go.ncsu.edu/hits) network (NSF award: [1730317](https://www.nsf.gov/awardsearch/showAward?AWD_ID=1730317&HistoricalAwards=false)). Our goal is to raise awareness of the use of high-throughput approaches and datasets using case study pedagogies.