General rules:

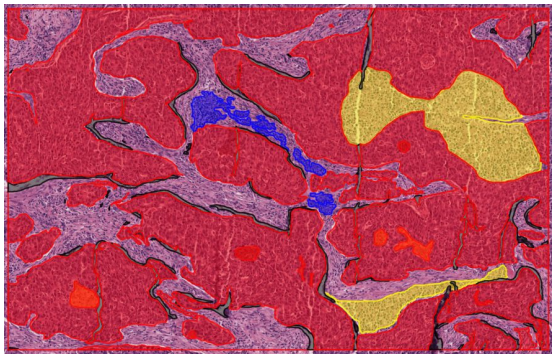
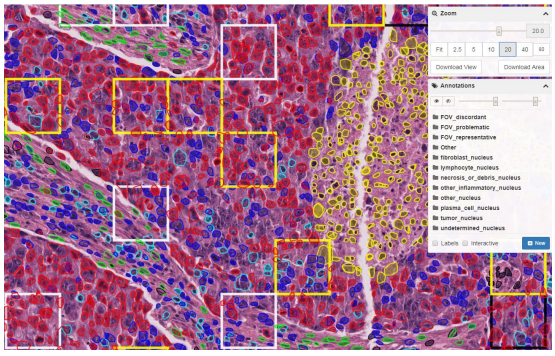
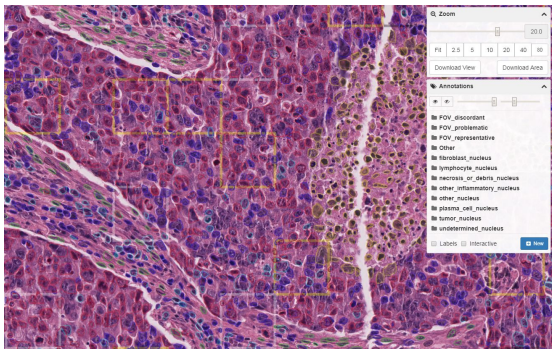
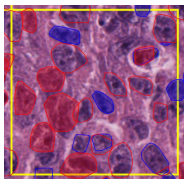
- Annotate **all** slides in folders associated with your name.
- Use a **comfortable mouse, table and monitor**. This will greatly impact comfort and work quality.
- When in doubt, take a screenshot of the nucleus and post it for review.
- Remember, the algorithm is **learning** what we teach it; poor quality annotations don't just hurt our chances of publication, but they also hurt everyone else who uses the data.

Specific annotation rules:

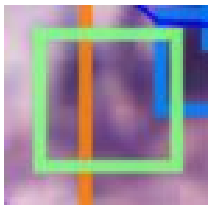
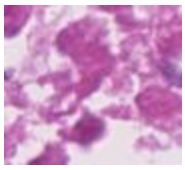
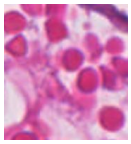
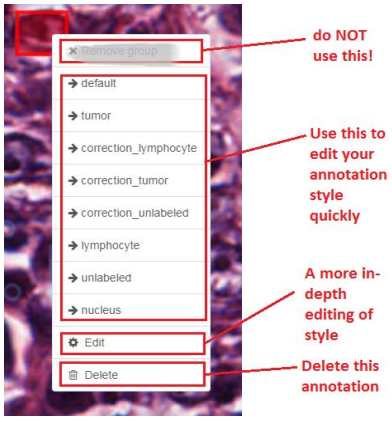
- Only annotate inside the predefined FOVs that were picked for you.
- If a nucleus extends beyond the FOV boundary, make sure your bounding box covers the full nucleus boundary (i.e. extend your rectangle outside the FOV as well).
- Make sure each FOV is fully complete before moving to the next FOV. It is very important to have high quality annotations. Missing annotations will confuse our algorithms!
- Nuclei are often vague. If you are unsure about the label of a nucleus, either:
    - Ask and receive feedback.
    - Assign is the label "unlabeled". (accepted for nuclei that are very vague)

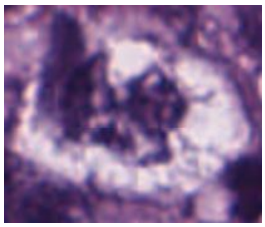
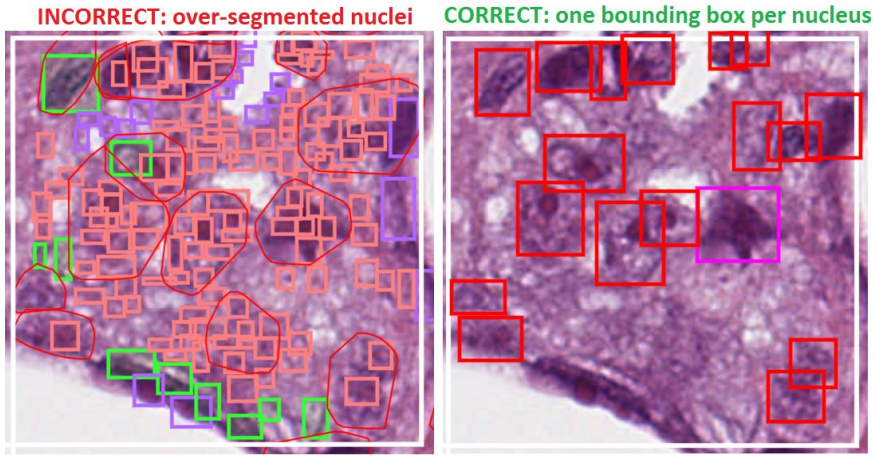
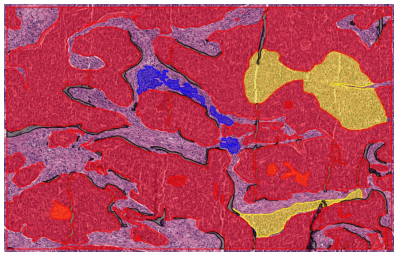
  Make as much effort to label nuclei as possible, only use the "unlabeled" label in a minority of cases.
- Make sure the bounding box is tight around the *nucleus* **NOT** the entire cell. Don't make bounding boxes that are too large.
- Never rotate the slide before annotating. All boxes should have the same orientation as the FOV. The slide should be in the same orientation as when it was first loaded.

- Annotation workflow (**VERY IMPORTANT**):

Step 1: Go to the same equivalent region annotations for the same slide in our published paper. Go to: https://goo.gl/cNM4EL Then scroll down and choose the slide. View the region annotations for that slide there. This will give you a general idea of what the various nuclei are. **This is especially relevant to knowing difficult classes like activated fibroblasts and endothelial cells.** In the image on the right (TCGA-A2-A0YM), everything not in the red regions is NOT tumor, so this is a big hint on how you should annotate your nuclei.	
Step 2: Now go back to that slide. Go to medium power (20X) and find the FOV you want to annotate. Reduce fill transparency (left bar) to zero.	
Step 3: Reduce the transparency of the boundaries themselves (right bar) and take a good look at the underlying tissue itself. You may take a look at the region annotations from step 1 if you are in doubt.	
Step 4: Zoom on the FOV itself at maximum power (40X or 80X, depending on slide) and start annotating. Continuously use the right bar to reduce and increase the boundary transparency as you need to see the tissue versus the annotations themselves.	

Illustrated rules + troubleshooting:

#	Situation	How to handle	Examples
1	A nucleus extends beyond the edge of the FOV and I need to place a rectangle	Extend your rectangle to encompass the full extend of the nucleus	 
2	I cannot see the underlying tissue properly	Reduce the fill transparency (left slider) or, less recommended, reduce the entire transparency (right slider)	
3	There is necrotic debris or collagen	Ignore it. Do **not** annotate debris or non-nuclear material.	 
4	There are red blood cells	Ignore. Do **not** annotate RBCs	
5	I would like to delete/edit my own annotations	You can edit or delete your annotations by right clicking on it. If you want to select multiple annotations, press and hold down the "control" key, then left click all annotations you want to select one at a time, while continuing to press on the control key. When you right click on the annotation or annotation group, you see a context menu that looks something like the image on the right.	

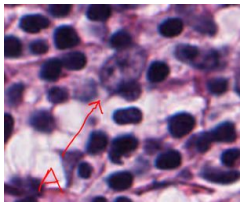
6	There is a multinucleated giant cell or a cell-eat-cell phenomenon (cannibalism)	Label each nucleus independently. We operate at the level of nuclei, not cells, in this project.	
7	There are overlapping nuclei and the bounding boxes will have to overlap to capture full extent.	No problem; use overlapping bounding boxes in this case.	
8	The nuclei are very textured and have prominent nucleoli	Don't be fooled!! Malignant nuclei can have a very textured appearance and prominent nucleoli so you may think they are multiple nuclei but are one nucleus!! By the way, in the image below, there are many vacuoles that were mistaken as being nuclei. This is a vacuolated phenotype. 	
9	The slide is quite difficult; stroma is difficult to distinguish from tumor.	Make sure you follow step 1 in the "Annotation workflow" table above. I.e. Make sure you go to https://goo.gl/cNM4EL and check the tumor regions there. Anything outside these regions may still be a tumor nucleus, but is more likely to be a fibroblast, lymphocyte, plasma cell etc.	

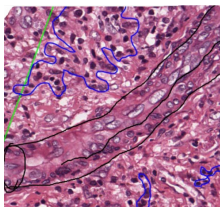
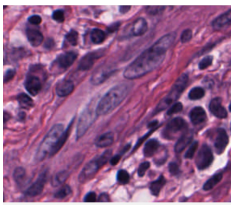
Standard styles to annotate:

***Note**: the styles only need to be imported once and they will be stored in your browser. It is important that you use the imported standard styles as-is. Please read through the following descriptions and notes before you start annotating.*

Style name	Description and Notes
tumor	Tumor nuclei are very heterogeneous in shape. They tend to have hyperchromatic, eccentric nuclei, and tend to be crowded and irregular. See any standard pathology textbook.
lymphocyte	Small, round, condensed, central nucleus. Tend to be grouped together.
plasma_cell	May be confused with lymphocytes. Plasma cells are much rarer than lymphocytes; when in doubt, ask the group. Tend to have more eccentric nucleus, more eosinophilic cytoplasm, a larger, textured nucleus (described as “cart-wheel”, but rarely seen as such) with a pale perinuclear halo.
fibroblast	Stromal nuclei that tend to be elongated and shaped like a cigar. May also have a rounder shape. The tell-tale sign is their present in stroma in alignment with the collagen fibres. Note: some tumor-associated fibroblasts become more “tumor-like” in morphology, also known as “activated fibroblasts”. When in doubt, take a screenshot of FOV and ask on Slack**.** *Habiba’s description: Fibroblasts (have elongated/spindle/ovoid dark nucleus with a dense eosinophilic tails that have the same intensity and color of the nearby stromal collagen, these tails are collages.*
unlabeled	If you are confident in your ability (eg you are a pathologist or have been annotating many FOVs), i.e. the nucleus is vague and cannot be labeled using just H&E, use this label.

Other styles (need pathologist feedback):

Style name	Description and example
mitotic_figure	
macrophage	 *Larger than neutrophils /lymphocytes, vacuolated or frothy cytoplasm (scant to moderate), thin round to reniform (bean shaped) nuclei with thin nuclear membrane that is central or eccentric, chromatin fine to coarse but uniformly distributed, nucleoli variable.*

endothelial_cell	 
ductal_epithelium	
myoepithelium	
apoptotic_body	
neutrophil	