

Submission ID #: 70973

Scriptwriter Name: Poornima G

Project Page Link: <https://review.jove.com/account/file-uploader?src=21646813>

Title: An Optimized Method to Isolate Female Mouse Urethral Epithelium for Single-Cell and Immunofluorescence Analysis

Authors and Affiliations:

AUTHORS AND AFFILIATIONS:

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Author Questionnaire

1. Microscopy: Does your protocol require the use of a dissecting or stereomicroscope for performing a complex dissection, microinjection technique, or something similar? **Yes , all done**

SCOPE: 3.1.2, 3.2.1, 3.7.1, 3.7.2, 4.1.1

2. Software: Does the part of your protocol being filmed include step-by-step descriptions of software usage? **No**

3. Filming location: Will the filming need to take place in multiple locations? **No**

4. Testimonials (optional): Would you be open to filming two short testimonial statements **live during your JoVE shoot**? These will **not appear in your JoVE video** but may be used in JoVE's promotional materials. **No**

Current Protocol Length

Number of Steps: 25

Number of Shots: 57 (4 Scope)

Interviews

Videographer: Obtain headshots for all authors available at the filming location.

Authors: Please note that **the questions will not appear on screen**. Answers are slightly edited as per journal guidelines and the best 5 answers have been retained. *Please answer in **stand-alone full sentence with sufficient context***

INTRODUCTION:

~~REQUIRED: Summarize your research focus and the main questions it aims to answer.~~

- 1.1. **Diya Binoy Joseph:** Our group is interested in epithelial patterning in the genitourinary tract. This protocol represents an optimized method to isolate the epithelial lining of the female mouse urethra for molecular and cellular studies.
 - 1.1.1. INTERVIEW: Named talent says the statement above in an interview-style shot, looking slightly off-camera

~~Describe the research contexts or systems in which this protocol can be applied.~~

- 1.2. **Diya Binoy Joseph:** The methods described in this paper will aid in research on the female urethral lining and its role in immune defense.
 - 1.2.1. INTERVIEW: Named talent says the statement above in an interview-style shot, looking slightly off-camera

CONCLUSION:

~~What does this protocol allow researchers to study or measure?~~

- 1.3. **Mrinal Samtiya:** This protocol allows researchers to study urethral epithelial cell types and self-renewal capacity of epithelial cells from the proximal and distal urethra.
 - 1.3.1. INTERVIEW: Named talent says the statement above in an interview-style shot, looking slightly off-camera

~~Describe the most important consideration or challenge when performing this protocol.~~

- 1.4. **Mrinal Samtiya**: One of the most important considerations of this protocol is the careful dissection of the full-length urethra and optimal enzymatic digestion to obtain viable urethral epithelial cells for downstream analyses.

1.4.1. INTERVIEW: Named talent says the statement above in an interview-style shot, looking slightly off-camera

~~Are there any additional methods or analyses that can be performed following this procedure?~~

- 1.5. **Mrinal Samtiya**: The epithelial cells produced by this protocol can be used for multiple applications, including single-cell RNA-sequencing, bulk RNA sequencing, flow cytometry, immunostaining, and organoid culture.

1.5.1. INTERVIEW: Named talent says the statement above in an interview-style shot, looking slightly off-camera

Videographer: Obtain headshots for all authors available at the filming location.

Ethics Title Card

This research has been approved by the Institutional Animal Ethics and the Institutional Biosafety Committee at the BRIC-Institute for Stem Cell Science and Regenerative Medicine

Protocol

***Videographer:** Please always record an audio slate while filming each shot mentioned in the shotlist.*

Videographer's NOTES:

Please refer to this file for all the shot numbers and their corresponding file names. Very helpful to AE: There are tags as "use this" to use a particular take for many shots

https://review.jove.com/files/ftp_upload/70973/JoVE_shoot-70973_25th-May-2026_Notes.pdf

1. Shot numbers are audio cues in the file. Shot numbers with corresponding file numbers are given in the above file for convenience for logging. Shots are not necessarily in order of script. Please check shot number.

2. Sometimes multiple takes were done. The best takes are indicated below with the comment USE THIS. Also indicated by CAN USE THIS when there are options.

3. Everything shot in 1080p. Shot at 25fps.

2. Isolation of the Adult Female Mouse Urethra

Demonstrator: Jasmine

Authors: During filming, please point out the organs or veins before dissecting them.

2.1. To begin, place the euthanized female CD-1 mouse, 6 to 12 weeks of age, in a supine position on the dissection table [1-TXT]. Spray 70% ethanol on the abdomen to wet the hair [2].

2.1.1. WIDE: Talent placing the euthanized mouse in a supine position on the dissection surface. **TXT: Euthanasia: CO₂ overdose followed by cervical dislocation** ***Videographer:** Please obtain a WIDE and CU shot as well for this shot.*

2.1.2. Talent spraying 70% ethanol on the abdominal hair of the mouse.

2.2. Using forceps, lift the skin on the abdomen [1]. Then, with small scissors, make a vertical incision toward the lower body, ending at the external urethral opening [2].

2.2.1. Talent lifting the abdominal skin of the mouse with forceps. **NOTE: 2.2.1 + 2.2.2 Combined shot : use DSCF0019**

- 2.2.2. Talent making a vertical incision along the abdomen toward the external urethral opening.
- 2.3. Identify the bladder and gently lift it with forceps [1]. If the bladder is filled with urine, gently compress it between the forceps to empty the bladder [2].
 - 2.3.1. Talent pointing to and lifting the bladder with forceps. **NOTE: 2.3.1 + 2.3.2 Combined shot : use DSCF0020**
 - 2.3.2. Talent gently compressing the bladder with forceps to empty the urine.
- 2.4. Using scissors, trim the fat and connective tissue around the bladder to expose the butterfly-shaped structure where the two pubic bones join at the pubic symphysis [1-TXT]. Observe the urethra running beneath the pubic symphysis [2].
 - 2.4.1. Talent trimming fat and connective tissue around the bladder using scissors. **TXT: Use small spring scissors for the entire protocol**
 - 2.4.2. Shot of pointing to the urethra running beneath the pubic symphysis.
- 2.5. Then, lift the pubic symphysis with forceps and cut it open using scissors [1]. Remove the pubic bone fragments to expose the thin urethra lying on the ventral surface of the vaginal wall [2].
 - 2.5.1. Talent lifting and cutting the pubic symphysis using forceps and scissors.
 - 2.5.2. Talent removing pubic bone fragments and pointing to the exposed urethra.
- 2.6. Using fine forceps, gently pull the bladder upward from the body cavity [1]. Run scissors between the urethra and the vaginal wall to separate the urinary and reproductive tracts [2].
 - 2.6.1. Talent gently pulling the bladder upward using fine forceps. **NOTE: 2.6.1 + 2.6.2 + 2.7.1 + 2.7.2 COMBINED DSCF0024**
 - 2.6.2. Talent separating the urethra from the vaginal wall using scissors.
- 2.7. Maintain slight tension on the bladder while cutting along the back of the urethra to separate it from the vaginal wall [1]. Continue cutting to the distal urethral opening to obtain the full-length mouse urethra measuring 1.2 to 1.5 centimeters in length [2]. Next, separate the urethra from the bladder by making a single cut at the bladder neck [3].

- 2.7.1. Talent cutting along the back of the urethra while maintaining tension on the bladder.
- 2.7.2. Talent continuing the dissection to isolate the full-length urethra. **Note: 2.7.2 additional shot DSCF0025 Use this. Ignore voice cue, refer to shot number here**
- 2.7.3. Talent making a single cut at the bladder neck to separate the urethra from the bladder.

3. Isolation of Urethral Epithelium from the Urethra

Demonstrator: Mrinal Samtiya

- 3.1. Pool 4 to 5 urethras in a sterile Petri dish containing fresh HBSS **[1-TXT]**. Under a dissecting microscope, trim the fat and connective tissue surrounding the urethra using fine-point forceps and scissors **[2]**.
 - 3.1.1. Talent transferring 4 or 5 dissected urethras into a sterile Petri dish containing HBSS. **TXT: HBSS: Hanks' Balanced Salt Solution; Use HBSS without calcium and magnesium throughout the procedure**
 - 3.1.2. SCOPE: 70973_screenshot_1.mp4 00:00-00:10.
- 3.2. Then, with scissors, cut each urethra crosswise into two equal-sized pieces **[1]**. Place the tissue pieces on top of a 100-micron nylon mesh strainer **[2]**.
 - 3.2.1. SCOPE: 70973_screenshot_1.mp4 00:11-00:15.
 - 3.2.2. Talent placing the urethra pieces onto a 100-micron nylon mesh strainer.
- 3.3. Wash the tissue pieces with 1 milliliter of HBSS **[1]** and transfer them into a 1.5-milliliter microcentrifuge tube containing 1 milliliter of 1% trypsin prepared in supplemented HBSS with antibiotics and antimycotics **[2]**. Incubate the sample on a rotator at 30 revolutions per minute and 4 degrees Celsius for 15 to 30 minutes **[3]**.
 - 3.3.1. Talent washing the tissue pieces on the nylon mesh strainer with 1 milliliter of 1x HBSS.
 - 3.3.2.** Talent transferring tissue pieces into a 1.5-milliliter microcentrifuge tube containing 1% trypsin solution. **NOTE: 3.2.2 + 3.3.1 combined shot**
 - 3.3.3. Talent placing the microcentrifuge tube onto a sample rotator for incubation.

- 3.4. After digestion, add magnesium chloride prepared in HBSS to the tube, followed by DNase I (1) prepared in PBS [1-TXT]. Incubate the tube for 15 to 30 minutes on a rotator placed inside a 37 degrees Celsius hybridization oven [2].
- 3.4.1. Talent adding magnesium chloride and DNase I solutions into the same tube.
TXT: Final concentrations: MgCl₂: 10 mM; DNase I: 10 µg/mL
- 3.4.2. Talent placing the microcentrifuge tube on a sample rotator placed inside a 37 degrees Celsius hybridization oven.
- 3.5. To quench the trypsin, transfer the urethral tissues into a 1.5-milliliter microcentrifuge tube containing 10% sheep serum in HBSS at room temperature [2].
- 3.5.1. Talent transferring urethral tissues into a microcentrifuge tube containing 10% sheep serum solution.
- 3.5.2. Talent placing the tube aside for room-temperature incubation.
- 3.6. After 5 minutes, transfer the urethral tissues into a new tube containing ice-cold HBSS [1] and then into a sterile Petri dish containing ice-cold HBSS [2].
- 3.6.1. Talent transferring the urethral tissues into a new tube containing ice-cold buffer.
- 3.6.2. Talent transferring the tissues into a sterile Petri dish containing ice-cold buffer.
- 3.7. While viewing the sample under a microscope, hold one end of the urethral piece with a pair of forceps [1]. Using another pair of forceps, pass along the entire length of the urethral piece to extrude the transparent epithelial sheet [2].
- 3.7.1. SCOPE: 70973_screenshot_2.mp4 00:00-00:04.
- 3.7.2. SCOPE: 70973_screenshot_2.mp4 00:05-00:14.
- 3.8. Process some of the isolated urethral epithelial sheets directly for whole-mount immunostaining to visualize epithelial cells and epithelial-associated macrophages in situ [1].
- 3.8.1. Talent transferring isolated urethral epithelial sheets into a well of a plate and adding antibodies. **Authors: Label the well-plate as "Whole-mount immunostaining".**
- Authors: This shot is placed here to maintain a good flow. It is only a representative shot to bridge the results and provide context. You do not need**

to perform the entire procedure.

4. Preparation of Single-Cell Suspension from Urethral Epithelium

Demonstrators: Mrinal Samtiya, Richelle Rodrigues

- 4.1. Transfer the epithelial pieces isolated from 4 to 5 pooled mouse urethras [1] into a sterile Petri dish with a small plastic transfer pipette [2]. Using scissors, mince the epithelium into small fragments in a drop of HBSS added directly to the tissue fragments [3].

Added shot: SCOPE: 70973_screenshot_3.mp4 00:00-00:05

- 4.1.1. Talent transferring isolated epithelial pieces into a sterile Petri dish using a transfer pipette. **NOTE:** Use DSCF0042 and DSCF0043
 - 4.1.2. Talent mincing the epithelial tissue into small fragments using scissors in a drop of buffer.
- 4.2. Transfer the minced epithelial fragments into a 2-milliliter microcentrifuge tube containing 1.5 milliliters of pre-warmed Accutase digestion solution at 37 degrees Celsius [1-TXT]. Incubate the sample on a rotator at 37 degrees for 1 hour [2].
 - 4.2.1. Talent transferring minced epithelial fragments into a microcentrifuge tube containing pre-warmed Accutase digestion solution. **TXT: Digestion solution: 1x Accutase; 10 μ M Rocki Y-27632; 100 μ g/mL DNase I**
 - 4.2.2. Talent placing the microcentrifuge tube onto a sample rotator inside the hybridization oven.
- 4.3. After incubation, transfer the entire contents into a 5-milliliter centrifuge tube containing 2 milliliters of wash buffer [1-TXT]. Centrifuge the suspension at 600 g for 5 minutes at 4 degrees Celsius using a swinging bucket rotor [1].
 - 4.3.1. Talent transferring the digestion mixture into a 5-milliliter centrifuge tube containing 2 ml wash buffer. **TXT: Wash buffer: 1x PBS without Ca and Mg + 1% BSA** **Authors:** Better to already have the wash buffer in the 5 ml tube and then add the tissue mix. This will save 1 shot as we are already at the 55 limit.
 - 4.3.2. Talent placing the centrifuge tube into a centrifuge with a swinging bucket rotor.
- 4.4. Then, aspirate the supernatant using a gentle vacuum [1] and resuspend the pellet in 1

milliliter of wash buffer with a 1-milliliter pipette with a filter tip [2].

4.4.1. Talent aspirating the supernatant from the centrifuge tube using a gentle vacuum.

4.4.2. Talent resuspending the pellet in wash buffer using a 1-milliliter pipette with a filter tip. **NOTE: 4.4.2 + 4.5.1 combined**

4.5. Using a fresh pipette tip, break up the pellet by pipetting up and down until the clumps become smaller [1]. Pass the suspension through a 23-and-a-half-gauge needle, then through a 26-and-a-half-gauge needle, each attached to a 1-milliliter syringe, using 4 strokes for each needle [2].

4.5.1. Talent pipetting the cell suspension up and down to break up the pellet.

4.5.2. Talent passing the cell suspension through a 23 1/2-gauge needle attached to a 1-milliliter syringe in 4 strokes.

4.6. Next, add 1 milliliter of wash buffer to wet the surface of a 30-micron pre-separation filter placed over a 5-milliliter tube [1]. Using the syringe connected to the 26-and-a-half-gauge needle, transfer 1 milliliter of the cell suspension through the filter [2].

4.6.1. Talent adding wash buffer onto the surface of a 30-micron pre-separation filter positioned over a 5-milliliter tube. **NOTE: 4.6.1 + 4.6.2 combined: DSCF0055 Use this. Please trim the middle of this shot to avoid the long gap between visible actions**

4.6.2. Talent passing the cell suspension through the pre-separation filter using the syringe and needle assembly.

4.7. Then, wash the tube with 1 milliliter of wash buffer and pass it through the filter [1]. Pellet the cells by centrifugation at 600 g for 5 minutes at 4 degrees Celsius using a swinging bucket rotor [2].

4.7.1. Talent adding wash buffer to the tube and then adding it on the filter.

4.7.2. Talent placing the tube into a centrifuge with a swinging bucket rotor.

4.8. Aspirate the supernatant [1]. Resuspend the pellet in 100 microliters of wash buffer and add 100 microliters of red blood cell lysis buffer [2]. After exactly 3 minutes, add 4 milliliters of wash buffer and mix [3]. Harvest the cells by centrifugation at 600 g for 5 minutes at 4 degrees Celsius using a swinging bucket rotor [4].

4.8.1. Talent aspirating the supernatant from the centrifuge tube.

- 4.8.2. Talent resuspending the pellet in wash buffer and adding red blood cell lysis buffer.
- 4.8.3. Talent picking up the tube from the rack and adding wash buffer and mixing.
- 4.8.4. Talent placing the tube into a centrifuge with a swinging bucket rotor.
- 4.9. After removing the supernatant, resuspend the pellet in 50 to 200 microliters of wash buffer according to the pellet size [1]. Mix a 5-microliter aliquot of cells with 5 microliters of trypan blue [2] and load the mixture onto a hemocytometer to assess cell yield and viability [3].
 - 4.9.1. Talent aspirating the supernatant and resuspending the cell pellet in wash buffer by pipetting up and down.
 - 4.9.2. Talent adding a 5-microliter aliquot of cells into a 0.5-milliliter microcentrifuge tube containing trypan blue and mixing.
 - 4.9.3. Talent loading the stained cell suspension onto a hemocytometer.

Authors: The following are representative shots to bridge the results and provide context. You need not perform the whole procedure.

- 4.10. Finally, perform fluorescent immunostaining to label epithelial cells, murine macrophages, and immune cells using appropriate antibodies [1]. Assess the cell viability and quantify distinct cell populations using flow cytometry [2].
 - 4.10.1. Talent adding fluorescent antibody solution to single-cell suspensions in centrifuge tube.
 - 4.10.2. Talent loading the sample into a flow cytometer.
- 4.11. Culture urethral epithelial single cells in a basement membrane matrix and organoid expansion media to generate three-dimensional urethral epithelial organoids [1]. Perform hematoxylin-eosin staining and fluorescent immunostaining on embedded organoid sections to visualize organoid structure and cellular composition [2 and 3].
 - 4.11.1. Talent dispensing epithelial cell suspension mixed with the basement membrane matrix into a multi-well plate. **Videographer's NOTES: DSCF0072 Use this as first half of shot 4.11.1 and use DSCF0073 Use this as second half of shot 4.11.1**
 - 4.11.2. Talent transferring organoid sections onto microscope slides and adding a stain from a labelled container.

Added shot 4.11.3 DSCF0070

Results

5. Results

- 5.1. The isolated urethral epithelial sheets had a higher transparency than [1] the stromal tissue after separation [2].
 - 5.1.1. LAB MEDIA: Figure 2I. *Video editor: Highlight the tissue labeled "Urethral epithelia"*
 - 5.1.2. LAB MEDIA: Figure 2I. *Video editor: Highlight the darker tissue labeled "Urethral stroma"*
- 5.2. Whole-mount immunostaining of the epithelial sheets showed strong labeling with the E-cadherin CDH1 antibody [1].
 - 5.2.1. LAB MEDIA: Figure 5A and C.
- 5.3. Labeling the sheets with the antibody to F4/80 (*F-four-eighty*) showed the macrophages forming branching dendritic structures between the epithelial cells of the urethra [1].
 - 5.3.1. LAB MEDIA: Figure 5E–F. *Video editor: Zoom in on the red cell extensions located between the green epithelial cell borders.*
- 5.4. Single cells isolated from the urethral epithelium showed consistently high viability by trypan blue exclusion on a hemocytometer [1] and flow cytometry using the amine-reactive ghost dye V450 (*4-fifty*) [1].
 - 5.4.1. LAB MEDIA: Figure 2L. *Video editor: Highlight the white round cells. DO NOT highlight the blue cells.*
 - 5.4.2. LAB MEDIA: Figure 3A. *Video editor: Show only the left panel labeled "Urethral epithelium singlets" and Highlight the gated region labeled "Live 83.09%".*
- 5.5. The isolated cells contained an enriched epithelial cell population after gating on the live cells [1].
 - 5.5.1. LAB MEDIA: Figure 3A. *Video editor: Show the 2 plots labeled "Urethral epithelium-Live" and Highlight the region upper left quadrants showing 92.16% and 90.57%".*
- 5.6. Approximately 1.3% of the CD45-positive immune cells and 1.18% macrophages were observed in the single cell suspensions [1].

- 5.6.1. LAB MEDIA: Figure 3A. *Video editor: Show the middle plot labeled “Urethral epithelium-Live” and highlight the region labeled “Immune-1.30%”.*
- 5.6.2. LAB MEDIA: Figure 3A. *Video editor: Show the extreme right plot labeled “Urethral epithelium-Live” and highlight the region labeled “Mac-1.18%”.*
- 5.7. Single epithelial cells isolated from the urethra remained viable and formed organoids in a 3-dimensional matrix [1], with organoid growth observed up to Day 6 in culture [2].
 - 5.7.1. LAB MEDIA: Figure 4B–G. *Video editor: Show the sequence of bright-field images from Day 0 to Day 6.*
 - 5.7.2. LAB MEDIA: Figure 4G. *Video editor: Highlight the larger round organoids visible in the Day 6 image.*
- 5.8. Hematoxylin-eosin staining showed 2 distinct morphologies of the urethral epithelial organoids [1], including large hollow organoids [2] and smaller solid organoids [3].
 - 5.8.1. LAB MEDIA: Figure 4H and I
 - 5.8.2. LAB MEDIA: Figure 4H and I. *Video editor: Highlight the large hollow circular organoid in H.*
 - 5.8.3. LAB MEDIA: Figure 4H and I. *Video editor: Highlight the smaller solid pink organoids in I.*
- 5.9. Both the hollow and the solid urethral epithelial organoids were stratified [1] and expressed KRT5 and P63 in the basal layer [2]. They expressed KRT13 in the suprabasal and intermediate cell layers [3].
 - 5.9.1. LAB MEDIA: Figure 4K–L.
 - 5.9.2. LAB MEDIA: Figure 4K–L. *Video editor: Highlight the green and white stained images in KRT5 and P63 columns.*
 - 5.9.3. LAB MEDIA: Figure 4K–L. *Video editor: Highlight the KRT13 images.*
- 5.10. The organoids generated from the proximal urethra showed a higher proportion of hollow organoids [1], while the organoids from the distal urethra formed more solid spheres [2].
 - 5.10.1. LAB MEDIA: Figure 4OP. *Video editor: Highlight the “Hollow” section in the “Proximal” bar*
 - 5.10.2. LAB MEDIA: Figure 4OP. *Video editor: Highlight the “Solid” section in the “Distal” bar*

6. Urethra
Pronunciation link: <https://www.merriam-webster.com/dictionary/urethra>
IPA: /jʊˈri:θrə/
Phonetic Spelling: yoo·ree·thruh
7. Urethral
Pronunciation link: <https://www.merriam-webster.com/dictionary/urethral>
IPA: /jʊˈri:θrəl/
Phonetic Spelling: yoo·ree·thruhl
8. Supine
Pronunciation link: <https://www.merriam-webster.com/dictionary/supine>
IPA: /suːˈpaɪn/
Phonetic Spelling: soo·pyne
9. Ethanol
Pronunciation link: <https://www.merriam-webster.com/dictionary/ethanol>
IPA: /ˈeθəˌnɔ:l/
Phonetic Spelling: eth·uh·nawl
10. Urethral
Pronunciation link: <https://www.merriam-webster.com/dictionary/urethral>
IPA: /jʊˈri:θrəl/
Phonetic Spelling: yoo·ree·thruhl
11. Pubic Symphysis
Pronunciation link: <https://www.merriam-webster.com/dictionary/symphysis>
IPA: /ˈpjuːbɪk ˈsɪmfəˌsɪs/
Phonetic Spelling: pyoo·bik sim·fuh·sis
12. Ventral
Pronunciation link: <https://www.merriam-webster.com/dictionary/ventral>
IPA: /ˈvɛntrəl/
Phonetic Spelling: ven·truhl
13. Epithelium
Pronunciation link: <https://www.merriam-webster.com/dictionary/epithelium>
IPA: /ˌɛpəˈθiːliəm/
Phonetic Spelling: eh·puh·thee·lee·uhm
14. Petri Dish
Pronunciation link: <https://www.merriam-webster.com/dictionary/Petri%20dish>
IPA: /ˈpiːtri dɪʃ/
Phonetic Spelling: pee·tree dish
15. HBSS
Pronunciation link: No confirmed link found
IPA: /ˌɛɪtʃˈbiː ɛs ˈɛs/
Phonetic Spelling: aych·bee·ess·ess
16. Microcentrifuge
Pronunciation link: <https://www.merriam-webster.com/dictionary/centrifuge>
IPA: /ˌmaɪkroʊˈsentrəˌfjuːdʒ/
Phonetic Spelling: my·kroh·sen·truh·fyooj

- 17. Trypsin**
Pronunciation link: <https://www.merriam-webster.com/dictionary/trypsin>
IPA: /ˈtrɪpsɪn/
Phonetic Spelling: trip·sin
- 18. Antimycotics**
Pronunciation link: <https://www.merriam-webster.com/dictionary/antimycotic>
IPA: /ˌæntɪ maɪˈkɑːtɪk/
Phonetic Spelling: an·tee·my·kot·ik
- 19. Magnesium Chloride**
Pronunciation link: <https://www.merriam-webster.com/dictionary/magnesium>
IPA: /mægˈniːziəm ˈklɔːraɪd/
Phonetic Spelling: mag·nee·zee·uhm klor·ide
- 20. DNase**
Pronunciation link: <https://www.howtopronounce.com/dnase>
IPA: /ˈdiːn eɪs/
Phonetic Spelling: dee·en·ace
- 21. Hybridization**
Pronunciation link: <https://www.merriam-webster.com/dictionary/hybridization>
IPA: /ˌhaɪbrɪdəˈzeɪʃən/
Phonetic Spelling: hy·brid·uh·zay·shuhn
- 22. Immunostaining**
Pronunciation link: <https://www.howtopronounce.com/immunostaining>
IPA: /ɪˌmjuːnoʊˈsteɪnɪŋ/
Phonetic Spelling: ih·myoo·noh·stay·ning
- 23. Accutase**
Pronunciation link: <https://www.howtopronounce.com/accutase>
IPA: /ˈækjoʊ teɪs/
Phonetic Spelling: ak·yuh·tays
- 24. Centrifugation**
Pronunciation link: <https://www.merriam-webster.com/dictionary/centrifugation>
IPA: /ˌsentrəfəˈgeɪʃən/
Phonetic Spelling: sen·truh·fuh·gay·shuhn
- 25. Supernatant**
Pronunciation link: <https://www.merriam-webster.com/dictionary/supernatant>
IPA: /suːˈpɜːnətənt/
Phonetic Spelling: soo·pur·nuh·tuhnt
- 26. Resuspend**
Pronunciation link: <https://www.merriam-webster.com/dictionary/resuspend>
IPA: /ˌriːsəˈspend/
Phonetic Spelling: ree·suh·spend
- 27. Microliter**
Pronunciation link: <https://www.merriam-webster.com/dictionary/microliter>
IPA: /ˈmaɪkroʊ liːtər/
Phonetic Spelling: my·kroh·lee·ter

- 28. Hemocytometer**
Pronunciation link: <https://www.merriam-webster.com/dictionary/hemocytometer>
IPA: /ˌhi:məsərˈtɑ:mətər/
Phonetic Spelling: hee·moh·sy·tom·uh·ter
- 29. Fluorescent**
Pronunciation link: <https://www.merriam-webster.com/dictionary/fluorescent>
IPA: /flʊˈresənt/
Phonetic Spelling: floor·ess·uhnt
- 30. Flow Cytometry**
Pronunciation link: <https://www.merriam-webster.com/dictionary/cytometry>
IPA: /floʊ saɪˈtɑ:mətri/
Phonetic Spelling: floh sy·tom·uh·tree
- 31. Organoid**
Pronunciation link: <https://www.howtopronounce.com/organoid>
IPA: /ˈɔ:rgənɔɪd/
Phonetic Spelling: or·guh·noyd
- 32. Hematoxylin**
Pronunciation link: <https://www.merriam-webster.com/dictionary/hematoxylin>
IPA: /ˌhi:məˈtɑ:ksəlin/
Phonetic Spelling: hee·muh·tok·suh·lin
- 33. Stromal**
Pronunciation link: <https://www.merriam-webster.com/dictionary/stromal>
IPA: /ˈstroʊməl/
Phonetic Spelling: stroh·muhl
- 34. Macrophages**
Pronunciation link: <https://www.merriam-webster.com/dictionary/macrophage>
IPA: /ˈmækroʊˌfeɪdʒɪz/
Phonetic Spelling: mak·roh·fay·jiz
- 35. Dendritic**
Pronunciation link: <https://www.merriam-webster.com/dictionary/dendritic>
IPA: /dɛnˈdritɪk/
Phonetic Spelling: den·drit·ik
- 36. Viability**
Pronunciation link: <https://www.merriam-webster.com/dictionary/viability>
IPA: /ˌvaɪəˈbɪləti/
Phonetic Spelling: vy·uh·bil·uh·tee
- 37. Organoids**
Pronunciation link: <https://www.howtopronounce.com/organoid>
IPA: /ˈɔ:rgənɔɪdz/
Phonetic Spelling: or·guh·noydz
- 38. Stratified**
Pronunciation link: <https://www.merriam-webster.com/dictionary/stratified>
IPA: /ˈstrætəˌfaɪd/
Phonetic Spelling: strat·uh·fyd

39. KRT13

Pronunciation link: No confirmed link found

IPA: /keɪ ɑːr tiː θɜːrˈtiːn/

Phonetic Spelling: kay·ar·tee thur·teen

40. Suprabasal

Pronunciation link: <https://www.howtopronounce.com/suprabasal>

IPA: /ˌsuːprəˈbeɪzəl/

Phonetic Spelling: soo·pruh·bay·zuhl

41. Proximal

Pronunciation link: <https://www.merriam-webster.com/dictionary/proximal>

IPA: /ˈprɔːksəməl/

Phonetic Spelling: prok·suh·muhl

42. Distal

Pronunciation link: <https://www.merriam-webster.com/dictionary/distal>

IPA: /ˈdɪstəl/

Phonetic Spelling: dis·tuhl