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Title: Comparing Methods to Measure the Morphology of Corroded Steel Bars

Authors' responses: the Title of our script should have been as follows below

Applicability Analysis of Assessment Methods for Morphological Parameters of Corroded Steel Bar

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Authors: Please ensure that the author names, affiliations, and contact information are all correct.

Authors: Please fill out the **Procedure Highlights** and **Critical Steps** in the summary below.

A. Microscopy: Does your protocol involve video microscopy, such as filming a complex dissection or microinjection technique? (Y/N) **N**

B. Software: Does your protocol include detailed, step-by-step instructions involving computer-controlled instrumentation or other software? (Y/N) **N**

C. Procedure Highlights: Of the steps to be filmed, which will viewers benefit **most** from seeing? Please list **4-6** steps from this script by their step numbers (*e.g.* 2.1).

Step 2.1, step 2.8, step 2.12 and step 2.13.

D. Critical Steps: What is the single most difficult aspect of this procedure? Please list **1-2** steps from this script and briefly describe how you ensure success.

Step 2.1: When using vernier calipers to measure bar diameter, we have to ensure that the vernier caliper is perpendicular to the steel bar.

Solution: Draw a group of the parallel straight lines on a white paper at a spacing of 10 mm. Hold the steel bar perpendicular to the parallel lines on the paper with the rubber mud and make sure that the marks on the steel bar coincide with the straight lines on the white paper. Make the heads of the caliper jaws approach to the straight line on the white paper, before move the jaws to touch the bar surface for the measurement of bar diameter.

Step 2.12: The generation of the scanned graphics in the 3D scanning method.

Solution: Observe the steel bar before scanning, find the feature points, and connect the different parts of the reinforcement through the overlap of the feature points.

E. Filming: Will filming need to take place in multiple locations? (Y/N) **Y**

If yes, how far apart are the locations? About 2.5 km

Authors: You do not need to use the “track changes” function if you would prefer not to; please use whichever editing method you are most comfortable with.

DRAFT: DO NOT USE FOR FILMING

1. Introduction (Opening Author Interviews)

Authors: The video will open with brief statements by the authors about the goal and significance of this work. **The introduction is limited to 150 words in total**. Each author may give **one** statement in the introduction. Each statement is limited to **30** words.

- Please do not write in this space -

A. [Required] Interview Statements (Said by you on camera. Don't forget to smile!)

Authors: Please complete the following two statements about the significance of your protocol and fill in the names of the speakers. **Each author may give only one statement in the introduction** unless only one author will speak in the video.

Please limit each statement to **30** words. You may modify the prompts to suit your protocol.

1.1. Author A – Professor Dawang Li: This video aims to measure the surface morphology of a steel bar with particular reference to a corroded steel bar.

1.2. Author B – Miss Ping Li: We will demonstrate and evaluate five different methods, including mass loss, vernier caliper, drainage, 3D scanning and XCT methods on a steel bar

B. [Optional] Statements for Additional Authors (Said by you on camera. Don't forget to smile!)

Authors: These optional statements allow **additional authors** to help introduce your protocol. Please remember that **the introduction (sections A-C) cannot exceed 150 words** and that each author may give only **one** statement in the introduction.

Please limit each statement to **30** words. You may modify the prompts to suit your protocol.

1.3. Author C - Dr Yingang Du – The 3D scanning is the best method for measurement of the spatial variability of corrosion penetration on the surface of a corroded bar.

1.4. Author D - Mr Wei Ren – The obtained corrosion using 3D scanning method would enable engineers to evaluate the safety and service life of existing engineering structures in our society much more precisely and reliably.

Authors: Interview statements will be edited for length and clarity. Please note that you will discuss your protocol further in the **conclusion**. To explain the background or essential technique for one or two steps of your protocol, please see the optional **Author Protocol Interview** section.

C. [Conditional] Introduction of Demonstrator (Said by you on camera. Don't forget to smile!)

Authors: Please fill out this section **ONLY** if one or more demonstrators have **not** given a statement in sections A or B. Please limit this section to **30** words.

1.5. Miss Ping Li: Demonstrating the procedure will be Mr Huilai Han, a technician from my laboratory, Mr Shenglin Cui, a technician from HOLON Three-dimensional Technology(Shenzhen) Co.,Ltd. (Introduce additional demonstrators in the same way.)

1.5.1. Miss Ping Li speaks towards the camera, interview style.

1.5.2. Mr Huilai Han looks up from instrument and acknowledges the camera.

1.5.3. Mr Shenglin Cui looks up from workbench and acknowledges the camera.

ANNOTATION KEY FOR PROTOCOL AND RESULTS

Shots and other visuals are listed in chronological order under the step in which they will be shown. Visuals are matched to the voice-over script with the annotation **[#-TYPE]**, where **#** is the third digit of the three-digit visual number and **TYPE** is an abbreviation for the type of visual. The types are:

WIDE: Wide zoom (as seen from ~1 m away)
MED or **MED-Over shoulder**: Medium zoom; can be positioned from an over-the-shoulder perspective
CU: Close-up; for fine detail or manipulations of small devices
ECU: Extreme close-up (as seen from ~1 cm away)

SCOPE: Video microscopy filmed through microscope eyepieces with a "scope kit".
SCREEN: Screen capture footage generated by authors.
LM: Figures, tables, animations, or videos ('lab media') provided by authors prior to or during scripting. This is primarily used in the Results section.

TXT: A text overlay will be shown with this shot. The text is included with the shot description.

For example, **[1-MED]** in step 2.1 indicates that shot 2.1.1 should be filmed with medium zoom. Annotations are placed at the **end** of the voice-over segment to be illustrated by the corresponding shot. For more information, please see our video guide to the format of JoVE scripts.

Protocol (Spoken by voice talent at JoVE.)

Authors: This is the script of the instructions for your protocol. To ensure that filming lasts only one day, the protocol is restricted to **60** shots in approximately **30** steps. Each step is limited to **3** lines of voice-over text (excluding annotations). The video article focuses on the aspects of the procedure that benefit from visual demonstration, but all essential steps must be mentioned.

2. Initial Specimen Testing

2.1. To begin, mark a polished 500-mm-long, 14-mm-diameter steel bar in 10-mm increments. **[1-MED-Over shoulder]** Then, use zeroed vernier calipers to measure the diameter of the bar at the first mark, **[2-MED]** with the jaws gently touching the bar. **[3-CU]**

2.1.1. Talent places a ruler next to the bar and starts marking the bar in 10-mm increments.

2.1.2. Talent places the vernier calipers across the bar and gently closes the calipers on the bar.

2.1.3. A side view of the bar showing the jaws of the calipers gently touching the sides of the bar.

Authors: The results section indicates that the bar is initially marked and the diameters at each mark measured before the 30-mm sections are cut from each end. Is the bar indeed marked before it is cut?

Authors' response: Yes, it needs to be marked before cutting.

2.2. Perform three more measurements, rotating the calipers by 45° each time, for a total of four measurements at 45° intervals. **[1-CU-TXT]** Repeat this process for each mark on the bar. Average the diameters at each mark and calculate the cross-sections. **[2-MED-Over shoulder-TXT]**

2.2.1. Talent loosens the calipers, rotates them by 45°, and closes the calipers around the bar again. (**TEXT**: Measure diameter at 0°, 45°, 90°, 135°) (The zoom level should be wide enough to show the entire pair of calipers, including the scale.)

2.2.2. Talent places the vernier calipers across the second mark on the bar, closes the calipers around the mark, and looks at the vernier scale. (**TEXT**: See text for all equations.)

2.3. Then, cut 30 mm from each end of the bar to obtain two 30-mm-long bar specimens **[1-CU]** and one 440-mm-long bar specimen. **[2-MED]** Weigh each specimen horizontally on a digital electronic scale and record the readings. **[3-MED-Over shoulder]**

2.3.1. Talent places the two 30-mm specimens one by one on the workspace.

2.3.2. Talent places the 440-mm specimen on the workspace with the 30-mm specimen.

2.3.3. Talent places one 30-mm specimen horizontally on the scale and then writes down the reading.

Authors: Do you mark the left and right ends of the specimens when you cut them?

Authors' response: Yes.

Authors: Would you be able to demonstrate cutting the specimens?

Authors' response: Yes, we are able to. We have a professional technician ready.

2.4. Mark the 30-mm-long bar specimens at 10-mm intervals starting 5 mm from the left side. **[1-CU]** Determine the average diameter at each of these marks as previously described. **[2-MED-Over shoulder]**

2.4.1. With the ruler placed next to a 30-mm specimen, talent marks the specimen at 5, 15, and 25 mm from the left side.

2.4.2. Talent places the vernier calipers across one of the new marks, closes the calipers on the mark, and looks at the vernier scale.

Authors: Do you indeed **re-mark** the 30-mm-long specimens?

Authors' response: Yes. Two 30 mm bar specimens were marked at the points of 5 mm, 15 mm and 25 mm from their left ends with an interval of 10-mm.

Authors: Do you **re-measure** the diameter of the 440-mm-long specimen as well, or do you use the previous measurements?
Authors' response: Yes, in the test, the steel bar is cut before measured using 4 methods.

2.5. Next, set up a static-hydraulic electromechanical universal testing machine. **[1-MED]** Place a glass displacement cylinder under the machine head. **[2-MED]** Fill the cylinder with tap water to just reach the outlet. **[3-CU]**

2.5.1. ~~Talent attaches the EUT head to the machine~~ Talent sets up the EUT instrument.

2.5.2. Talent places the glass container under the EUT head.

2.5.3. Talent fills the container with tap water until the water level just reaches the outlet.

Authors: Do you usually need to attach the EUT head? What else (if anything) do you need to do to set up the machine?

Authors' response: No! The EUT comes with a head and does not need to attach it. Hence, the step 2.5.1 can be modified as the above.

2.6. Then, place an electronic scale below the outlet of the container. **[1-MED]** Place a 200-mL beaker on the scale in line with the outlet. **[2-MED-Over shoulder]**

2.6.1. Talent places the scale next to the container on the outlet side.

2.6.2. Talent puts the beaker on the scale under the outlet.

Authors: Do you zero or tare the scale?

Authors' response: Yes! The reading of the electronic scale needs to be zeroed, before the start of the measurement.

2.7. After that, clamp one end of a bar specimen vertically in the head of the EUT machine. **[1-MED]** Move the EUT head down vertically **[2-MED]** until the other end of the bar just touches the surface of the water. **[3-CU]**

2.7.1. Talent clamps a bar specimen in the EUT machine.

2.7.2. Talent uses the handset/console to send the EUT head down to move the bar towards the water.

2.7.3. A side-on view of the water surface as talent moves the bar down by the last 1-2 mm to just touch the water.

Authors: How much of the bar is taken up by the grip of the EUT head?

Authors' response: According to the instructions of the EUT, the steel bar caught should occupy 3/4 of the head length.

Authors: Which bar specimen do you plan to use for this demonstration? (I have set the zoom levels here for the 440-mm specimen, but I can change them if you would prefer to use a 30-mm specimen.)

Authors' response: 440mm steel bar will be used only for this demonstration.

Authors: How far above the water is the end of the bar specimen when you first clamp it into the machine? How quickly do you move the bar down to the water initially when you are directly controlling it? (This just affects the pacing of the second and third shots.)

Authors' response: The bottom end of the clamped bar will be about 20mm above the water. The bar end will initially move down at 10mm per minute before touch water surface.

2.8. Record the initial reading on the digital scale. **[1-MED-Over shoulder]** Then, set the EUT machine to move the bar downward at 10 mm/min. Run the machine **[2-CU]** to displace the bar to the first 10-mm mark. **[3-MED]**

2.8.1. Talent checks the scale reading and writes down the displayed value.

2.8.2. A view of the handheld interface as talent sets the EUT machine to 10 mm/min and then starts the displacement.

2.8.3. The bar displacement stopping as the bar is submerged just to the mark.

Authors: Can you set the instrument to displace the bar by a certain distance/for a certain duration, or do you need to watch the bar and manually stop the machine?
Authors' response: Yes, the displacement of a steel bar into the water is controlled by the computer. It stops at each 10mm.

2.9. Then, record the final reading on the electronic scale. **[1-MED]** Calculate the uniform cross-section of the 10-mm segment of the bar from the mass of the displaced water. **[2-MED-Over shoulder]**

2.9.1. Talent looks at the scale after the displacement and writes down the "after" mass. The beaker containing the displaced water on the scale should be visible in this shot.

2.9.2. Talent enters the 'before' and 'after' masses into a spreadsheet (or uses another method) to calculate the cross-sectional area.

Authors: Do you use a spreadsheet to automatically calculate the cross-sectional area for each segment? If so, do you calculate the cross-sectional area for each segment as you measure it, or do you enter the values all together when you finish the test?

Authors' response: Yes. We use a spreadsheet to automatically calculate the cross-sectional area for each segment and enter the values all together when we finish the test.

2.10. Repeat this process for each 10-mm segment of the bar specimen **[1-MED]** until the entire bar is submerged. Test each of the three specimens in this way. **[2-MED]**

2.10.1. Talent starts the displacement for the next segment.

2.10.2. With the bar now almost completely submerged, talent stops the instrument (if it needs to be manually stopped) once the bar is completely submerged.

Authors: Do you let the bars air-dry after this test?
Authors' response: Yes, it will be dried in air for one hour.

2.11. Next, spray each bar specimen with white flaw detection developer and allow the specimens to dry in air. **[1-MED-Over shoulder]** Place one coated specimen on the platform of a 3D scanner. **[2-WIDE]**

2.11.1. Talent sprays the three bar specimens with white developer and then leaves them to dry.

2.11.2. Talent places the 440-mm specimen (or a 30-mm specimen, if preferred) under the 3D scanner.

Authors: Do you re-mark the bars at 10-mm increments once the spray has dried?
Authors' response: No. no mark is required for 3D and XCT methods

Authors: About how long does it take for the developer to dry? Do you spray the bars all over or do you spray just the top half of the bars, let them dry, turn over the bars, and spray the other side?
Authors' response: 1 to 2 minutes for the developer to dry. A steel bar will be spayed over its whole length before measured using 3D scanning and XCT methods.

2.12. Calibrate the bar specimen position using a label marked with a random array of small white dots. **[1-MED]** Then, scan the specimen along its length, develop a spatial model, and generate morphological data from the model. **[2-MED-Over shoulder]** Repeat this for each specimen. **[3-WIDE]**

2.12.1. Talent places the label around the bar specimen.

2.12.2. Talent starts the scan from the 3D scanner software and watches the scan proceed for ~5 s.

2.12.3. Talent removes the 440-mm specimen and places a 30-mm specimen under the 3D scanner.

Authors: Is the label pre-made, or do you need to make one (i.e., with the white paint pen on label paper) for each bar? Do you have a preferred background color for the label?

Authors: Where do you place the label relative to the bar?
Authors' response:
a) This is prepared beforehand.
b) There is no specific requirement for the color of the label. What we need is that the color can be clearly identified by the scanner. The scanned table is black, the steel bar is white, usually, we use a white label compared to the black background.
c) The steel bar is placed on the scanned table, and the labels can be stuck directly onto the table

2.13. Next, place one 30-mm-long specimen on the rotatable platform of an X-ray micro-computed tomography system. **[1-MED-Over shoulder]** Close the XCT instrument, open the instrument software, **[2-WIDE]** and adjust the specimen to the correct position for scanning. **[3-MED-Over shoulder]**

2.13.1. Talent opens the XCT instrument and places a 30-mm specimen on the platform.

2.13.2. Talent closes the instrument and goes to the XCT computer.

2.13.3. Talent adjusts the bar specimen to the shooting position.

2.14. Fill in the desired pixel size and magnification factor. Then, scan the specimen and generate the geometric parameters from this data. **[1-MED-Over shoulder]** Repeat the process for the other 30-mm-long specimen. **[2-MED]**

2.14.1. Talent sets the pixel size and magnification factor, and then starts the scan and monitors the scan for ~3-4 seconds.

2.14.2. Talent opens the XCT instrument door, removes the first 30-mm specimen, and places the second 30-mm specimen in the instrument.

Authors: Do you have any recommended parameters for the XCT?

Authors' response: The pixel size is 31.8121 μm and the optical magnification is 0.39964 in this test.

Authors' corrections: Generally, a sample shooting requires 2-4h (according to the set parameters) and can be not opened in the midway of a test.

3. Specimen Corrosion and Subsequent Testing

3.1. After acquiring the initial measurements of non-corroded steel bar, Attach an electrical wire to one end of the specimen. **[1-MED-Over shoulder]** Wrap 70 mm of each end of the 440-mm-long bar specimen in insulation tape. **[2-MED]**

3.1.1. Talent wraps one end of the bar in insulation tape.

3.1.2. Talent ~~starts wrapping tape around the other end of the bar, and~~ then holds a wire in place against the bar and tapes the wire against the bar.

Authors' corrections: Just wrap the wires directly, then wrap in insulation tape.

Authors: Do you need to remove the white developer before starting this process? If so, how do you remove it?

Authors' response: Yes, remove the white developer using with clean water.

Authors: Do you attach the wire by placing it against the side of the bar before wrapping it in tape? Do you have a preferred type of electrical wire? How long should the exposed part of the wire be? (i.e., what length of exposed wire should be in contact with the bar?) Do you have a recommended overall wire length or position along the bar where the wire should be attached?

Authors' response: Yes, we attached the wire by placing it against the side of the bar before wrapping it in tape. The type of electrical wire used in the test is BVR 2.5 mm^2 and with 50-60mm long exposed and contacted with the steel bar. The wire is about 2.0 m long and attached to one end of 440 mm bar.

3.2. Then, mix epoxy adhesive with hardener in a 1:1 ratio. **[1-MED]** Apply the epoxy uniformly to each insulated end of the bar to protect the ends from corrosion. **[2-MED/CU]**

3.2.1. Talent mixes together the epoxy adhesive and hardener.

3.2.2. With epoxy already having been applied to the end of the bar without the wire, talent applies epoxy to cover the end of the bar with the wire. (The zoom level should be wide enough to show the entire bar.)

Authors: How quickly does the epoxy set? If it sets very quickly, you may need to prepare a second batch of epoxy to film 3.2.2, as it takes a few minutes to set up between shots.
Authors' response: It may take 24 hours to set. But for better shooting, we will prepare an epoxy resin solidified steel in advance.

3.3. Once the epoxy has dried, fill a plastic tank equipped with a copper plate with an aqueous 3.5% solution of NaCl. **[1-WIDE]** Place the bar specimen in the tank**[2-MED]**

3.3.1. Talent pours the solution (or just water, if the solution is mixed in the tank) into the plastic tank (which, if the solution is mixed in the tank, already contains the needed NaCl).

3.3.2. Talent places the specimen in the tank.

Authors: Do you mix the solution in a separate container, or in the plastic tank? Should the solution depth be about 15 mm?
Authors' response: No, the solution will be mixed in the same tank and exceed 15mm.

Authors: How large is the copper plate? How is it attached to the plastic tank (if it is attached)?

Authors' response:
a) The cooper plate has a size of 400 mm x 100 mm
b) In the test, the copper plate was placed in parallel with the steel in the tank with some small wooden blocks stand, and there was no contact with each other

Authors: Is the electrolyte solution 3.5% by weight? What grade of water do you use for the solution? (e.g., deionized, ultrapure, etc.)

Authors' response: Yes, it is 3.5% salt electrolyte solution of tap water

3.4. Then, connect the wire attached to the bar to the positive terminal of a DC power supply. **[1-MED-Over shoulder]** Connect the copper plate to the negative terminal of the power supply. **[2-MED]**

3.4.1. Talent clips the wire from the bar to the positive terminal of the power supply.

3.4.2. Talent clips the wire from the copper plate to the negative terminal of the power supply.

Authors: How far is the power supply from the tank? (i.e., how far would the demonstrator need to move to connect the wires?)
Authors' response: In order to facilitate shooting, the relevant facilities will be gathered within an area of about 2.0 m^2.

Authors: Do you have a preferred type of electrical wire that you use for connecting the copper plate to the power supply?

Authors' response: Yes, we use the same wire as those for the steel bar.

Authors: Does the copper plate have a wire permanently connected to it, or do you need to attach a wire to it every time? If the wire is not permanently connected, do you attach the wire to the copper plate (with the other end disconnected) before filling the tank with the electrolyte?

Authors' response: No. a wire is connected to the copper plate before shooting.

Authors: Do you indeed use clips, or do you connect the wires by twisting them around the terminal? If you use clips, does the clip need to be connected to the other end of the wire attached to the bar before connecting the wire to the bar, or do you fit the wire with a clip afterwards?
Authors' response: No, the wires is directly twisted around the terminal in the test.

3.5. Set the power supply to produce a current density of 2.5 μA/cm^2 throughout the bar. **[1-MED-Over shoulder]** Apply the current for the duration needed to attain the desired level of corrosion per Faraday's law. **[2-CU]**

3.5.1. Talent sets the power supply to the needed current to produce 2.5 µA/cm^2 and turns on the power supply.

3.5.2. The bar specimen corroding in the tank. (5-7 s of footage)

Authors: Is this specifically the corrosion current density?
Authors' response: Yes.

Authors: Can you set the current before turning on the power supply at all (i.e., does it have analog controls)?
Authors' response: No, it will be set up after turning on the power.

3.6. Then, turn off the current **[1-MED-Over shoulder]** and transfer the bar specimen to a tank filled with a 12% solution of hydrochloric acid. **[2-MED]** Soak the bar in hydrochloric acid for 30 minutes to remove corrosion products. **[3-CU]**

3.6.1. Talent turns off the power supply.

3.6.2. Talent transfers the corroded bar to a labeled tank of HCl.

3.6.3. The bar soaking in the HCl tank. (5-7 s of footage)

Authors: Do you disconnect the bar before transferring it to the HCl?

Authors' response: Yes, of course!

Authors: Is the HCl solution percent by volume (% v/v)?

Authors' response: Yes, it is.

Authors: Do you use a tool to transfer the bar between the tanks? If so, what is it? Do you place the HCl tank next to the NaCl tank to make transferring the bar easier?

Authors' response:
a) The anti-acid gloves will be used for the transfer of the corroded steel bar from the salt tank to the acid tank.
b) Yes, both tanks are in the same place with a good ventilation.

3.7. After that, transfer the bar specimen to a saturated limewater solution to neutralize the acid residue. **[1-MED]** Then, rinse the bar with tap water and allow it to dry in air. **[2-WIDE]**

3.7.1. Talent removes the bar from the HCl and immerses it in the limewater tank.

3.7.2. Talent rinses the bar with tap water and places it aside to dry.

Authors: Do you dry the bar on a rack near the sink, or is the drying area further away?

Authors' response: It is dried in the area further away from the sink.

3.8. Next, mark the corroded surface at 10-mm intervals. **[1-MED-Over shoulder]** Weigh the corroded bar horizontally on a digital electronic scale and calculate the average area of the corroded bar. **[2-MED-Over shoulder]**

3.8.1. With a ruler already placed next to the corroded bar, talent marks the bar at 10-mm intervals.

3.8.2. Talent places the bar horizontally on the zeroed scale and writes down the reading once it stabilizes.

3.9. Determine the average diameter at each 10-mm mark using vernier calipers and calculate the cross-sections. **[1-MED]**

3.9.1. Talent places the calipers across one mark, carefully closes the calipers around the bar, checks the reading on the vernier scale, writes down the reading, and then adjusts the calipers by 45°.

3.10. Then, fix the bar in the EUT machine and calculate the cross-sections of each 10-mm segment of the corroded bar using the displaced water drainage method. **[1-MED]**

3.10.1. Talent secures the bar in the EUT machine, lowers the bar into position for the EUT test, and checks the starting weight in the beaker.

Authors: For the drainage method, should the test start with the 70-mm epoxy-coated section immersed (i.e., with the water just at the bottom of the corroded section) and end when the corroded section is fully immersed?

Authors' response: Yes, the test starts with the 70-mm epoxy-coated section immersed and ends when the corroded section is fully immersed.

3.11. After that, spray the specimen with white developer and take a 3D scan. **[1-WIDE]** Lastly, cut a 30-mm segment from the corroded bar and scan it with XCT. **[2-MED-Over shoulder]**

3.11.1. Talent places the corroded bar (now sprayed with developer) under the 3D scanner and goes to the computer to start a 3D scan.

3.11.2. Talent places a 30-mm segment of the corroded bar in the XCT instrument and closes the instrument door.

Authors: As some steps in your procedure have long reaction or waiting times, you may need to prepare the products of those steps in advance so that filming can easily move from step to step. Please see the **General Preparation** section at the end of this document for more information about preparing to demonstrate your protocol.

Authors' response: Yes, we will get to prepare the products of those steps in advance.

OPTIONAL: AUTHOR PROTOCOL INTERVIEWS

Authors: You may provide a brief statement about the single most critical step of the procedure. If there are two critical steps, you may provide two statements, but **this is the maximum**. Please limit each statement to **40** words.

This will be an **interview shot** following the listed step of the protocol. Please specify who will be speaking and the step that each statement should accompany.

Optional interview statement 1: Author Name, Step X.X: (Statement about step X.X)

Optional interview statement 2: Author Name, Step Y.Y: (Statement about step Y.Y)

RESULTS (Spoken by voice talent at JoVE; animated by video editor at JoVE.)

Authors: The representative results section presents the outcome and validation of your procedure. For example, syntheses would include product characterization and the reaction scope. Device or instrument fabrication and use would present examples of recorded data or products generated.

The results section is limited to **200** words. Please review the narration and proposed animations to ensure that they correctly summarize your results and appropriately highlight the relevance of your procedure.

4. Results: Comparison of Measurement Methods

4.1. The measured diameters of the intact steel bar did not vary significantly along its length, but consistent variation was observed between **[1-LM]** the 45° measurement and the 135° measurement, indicating that the bar was elliptical. **[2-LM]**

4.1.1. **Figure 6 (57859-Du and Wei-6A.tif)**: *Video editor*: On "consistent variation…", highlight the difference between the 45° line in 6A (red dashes) and the 135° line in 6A (black dashes).

4.1.2. **Figure 6 (57859-Du and Wei-6B.tif):** *Video editor*: Emphasize the 45° minimum and maximum bars in 6B. On "the 135°…", also emphasize the 135° bars in 6B.

4.2. The mass loss, caliper measurements, and 3D scanning techniques produced similar values with low variation. **[1-LM]** The drainage method measurements of the 440-mm-long specimen suffered from multiple sources of uncertainty, including the dryness of the bar and the surface tension of the water. **[2-LM]**

4.2.1. **Figure 7 (57859-Du and Wei-7.tif):** *Video editor*: Highlight the line of black squares (mass loss data) in both graphs when "mass loss" is spoken, the line of red dots (caliper data) in both graphs when "caliper measurements" is spoken, and the line of pink triangles (3D scanning data) in both graphs when "3D scanning" is spoken.

4.2.2. **Figure 7A (57859-Du and Wei-7A.tif):** *Video editor*: Emphasize the line of blue triangles (drainage data).

4.3. The 30-mm specimens were analyzed with XCT, which produced values consistent with the other techniques. **[1-LM]** Overall, calipers, XCT, and 3D scanning produced similar values with minimal variation. **[2-LM]** Thus, caliper measurements were the simplest way to obtain accurate measurements of non-corroded bars. **[3-LM]**

4.3.1. **Figure 7B (57859-Du and Wei-7B.tif):** *Video editor:* On "XCT", emphasize the line of green diamonds. On "consistent with…", emphasize the other four lines.

4.3.2. **Table 1** (Table 1.xls): *Video editor*: During "calipers", emphasize the 'Caliper method' column. During "XCT", emphasize the 'XCT method' column. During "3D scanning", emphasize the '3D scanning method' column.

4.3.3. **Figure 7 (57859-Du and Wei-7.tif)** and Table 1 (Table 1.xls): *Video editor*: Emphasize the 'Caliper method' column in Table 1 and the line of red dots in both panels of Figure 7 (the caliper data).

4.4. Corrosion resulted in substantial variation in cross-sectional shape throughout the bar, **[1-LM]** which could not be captured with the mass loss method. **[2-LM]** While calipers were more sensitive to the shape variation, they could not account for pitting in the surface of the specimen. **[3-LM]**

4.4.1. **Figure 8 (57859-Du and Wei-8.tif):** *Video editor*: Emphasize images 1, 2, and 3 in 8B (showing the varied cross-sections from the numbered locations in the picture to their left).

4.4.2. **Figure 9 (57859-Du and Wei-9.tif)**: *Video editor*: Emphasize the line of black squares (the mass loss data) in both graphs.

4.4.3. **Figure 9 (57859-Du and Wei-9.tif):** *Video editor*: Emphasize the line of red dots (the caliper data) in both graphs.

4.5. XCT and 3D scanning produced similar values, **[1-LM]** but XCT is limited by the need for small samples with flat ends. **[2-LM]** Thus, 3D measurements were favored for analyzing the morphology of corroded steel bars. **[3-LM]**

4.5.1. **Figure 9 (57859-Du and Wei-9.tif):** *Video editor*: Emphasize the line of pink triangles (3D scanning) in both graphs and the line of green diamonds (XCT) in 9B.

4.5.2. **Figures 8B (57859-Du and Wei-8B.tif) and 9B (57859-Du and Wei-9B.tif):** *Video editor*: Emphasize the left image in 8B (the image of an XCT sample).

4.5.3. **Figure 9 (57859-Du and Wei-9.tif):** *Video editor*: Emphasize the line of pink triangles in both graphs.

Authors: Please upload each panel of Figures 7, 8, and 9 as individual image files. Please remove the A/B labels from each of these images. See the **Provided Media** section for more information about uploading files for video inclusion.

Authors' response: Yes, each panel of Figures 7, 8, and 9 has been attached. The A/B labels have been removed from each of these images

Authors: Does the above description accurately reflect your results? See the instructions below to make changes.

Authors' response: Yes, they have.

Authors: The results section is limited to **200** words. All results described in detail must be illustrated. Please limit the narration to 3 lines per figure panel, table, or movie. Please remove 'Figure 1' and 'a)' labels from images and include scale bars and legends.

Please use **non-technical language** to describe areas of figures or tables to be highlighted during the voice-over. Please provide the pronunciation of technical terms (phonemic or IPA) for the JoVE voice talent.

Upload new and modified files to **your project folder**. Please upload each figure panel separately. File naming conventions and preferred file formats are described in the **Provided Media** section.

Each image, table, or movie will be referenced with its file name in the final script. Your script editor will ensure that the narration and animation descriptions are correctly formatted. For example:

5. Results: Evaluation of Morpholino Injection and Knockdown

5.1. Representative results of both morpholino injection and mRNA injection are shown here. The un-injected control at 48 h post-fertilization looks normal, as expected. **[1-LM]**

5.1.1. Figure 1 (01234_PIname_Figure1.tif): Non-technical description of areas of the figure to highlight.

5.2. However, embryos injected with the morpholino heg_e3i3_egfr1 (hegg E-3-I-3 E-G-F-R-1), which knocks down Heg isoforms containing the first of two EGF-like repeats, exhibit brain edema. **[1-LM]**

5.2.1. Figure 2 (01234_PIname_Figure2.tif)

5.3. Injection of heart of glass mRNA also produced an obvious phenotype. At 24 h post-fertilization, the heads of the un-injected controls look normal. **[1-LM]** Conversely, some of the embryos injected with the mRNA exhibit cyclopia. **[2-LM]**

5.3.1. Figure 3 (01234_PIname_Figure3.tif)

5.3.2. Figure 4 (01234_PIname_Figure4.tif)

To see how the narration is paired with figures, please visit http://www.jove.com/video/1597/results-example-mably?status=a3603k. File names are not shown in the final video article.

5. Conclusion (Said by you on camera. Don't forget to smile!)

Authors: The video will conclude with one or more author statements providing important reminders or discussing the scope of this technique. Please complete at least one statement below and identify each speaker. Please limit each statement to **40** words. You may modify the prompts to suit your protocol.

5.1. Author A – Professor Dawang Li: A vernier caliper is the best tool for measuring non-corroded steel bar's surface morphological, and it's easy to operate and costs most economically

5.2. Author B – Professor Yingang Du: The drainage method measured results may be affected by some uncertainties, and it is needed a further improvement on the measurement device.

5.3. Author C – Miss Ping Li: Although the XCT method can accurately measure the residual section area of a corroded steel bar, it is limited by the length of the specimen.

5.4. Author D – Mr. Ren Wei: The 3D scanning method is the most optimal method for the measurement of a corroded steel bar for its precision, economical and efficient, and it can generate more useful information of a corroded bar.

Authors: Statements will be edited for length and clarity. I am happy to help if you have questions.

PROVIDED MEDIA

Authors: Name new or modified files with the scheme 01234_PIname_Figure1.tif, where 01234 is your JoVE video ID and PIname is the corresponding author's surname. For example:

5.2 – *01234_PIname_Figure1.tif* – dual color imaging of tumor angiogenesis at 40X
5.3 – *01234_PIname_Figure2.tif* – dual color imaging of tumor angiogenesis at 100X

Minimum dimensions: 720 x 480 pixels
Minimum resolution: 300 dpi

Preferred image formats: .pdf, .tiff, .png, .eps, .ai, .psd
Preferred movie formats: .mov, .mp4, .avi

.mov or .mp4 files are strongly preferred for screen capture footage. Vector or layer-compatible formats (.svg, .ai, .eps, .pdf, .psd) are strongly preferred for complex figures and graphs.

To generate a vector (.pdf) file from a graph prepared in Excel or similar software, move the graph to its own sheet and save the sheet as a standard .pdf file.

Upload each file to your project folder: https://www.jove.com/account/file-uploader?src=17647043

Please list the provided files below and specify the step or steps where the files will be used. If a file is not based on an existing figure, please provide a short description.

- Step number(s) – *File name* - Description (if new figure)

Authors: If you have trouble uploading your files or if you have questions about preparing files for the video, please contact me at tara.cass@jove.com; I will be happy to help.

Back to Results

GENERAL PREPARATION

It is **critical** for a smooth and organized shoot that your samples, reagents, instruments, glassware, and software are ready to go. This ensures that filming can quickly move from step to step.

Reagents, samples, and solutions should be prepared or collected and labeled before we arrive. Tubes, glassware, and plates should be clean, dry, and neatly labeled.

Each shot will take about five minutes, as it takes a few minutes to set up between shots. You may need to prepare duplicate samples if a step that must be performed quickly is shown with more than one shot.

The filming process will be like a cooking show: if a step takes more than 5-10 minutes, you will continue the demonstration with the pre-made product of that step. Therefore, **if your procedure has**

long incubation, reaction, heating, or calculation times, please prepare the products of those steps before we arrive. Please notify your script editor if the product of a long step is too unstable to be prepared in advance.

Individual shots may be filmed out of order to allow a longer process to finish. If your procedure has a long delay between sample preparation and the experiment itself, you may need to film the experiment before filming sample preparation. Please clearly mark shots or steps that you wish to film out of order in the script.

Please contact your script editor or see JoVE's FAQ if you have general questions about filming. You will receive detailed preparation instructions in the email accompanying the finalized script.