Dear referee, dear editor,

Thank you again for the review, the comments were helpful in sharpening the message of the paper and were further encouraged by the positive feedback we received on the revision. We have also added the zenodo link to all the data and code used. Due to some technical issues on our end, some folders are out of date but the data is fully there. We are currently working on a solution and will update it as soon as we can.

In the below, the original reviewer comments are formatted in black normal font with green highlights, whereas our responses are formatted in red.

Editor Evaluations:
Recommendation: Accept Revision Required

Editor (Comments for the Author (Required)):

Please do not use the word "cavitation." Cavitation is the spontaneous phase change from liquid to vapor and is not how we understand hydraulic dysfunction to occur in plants. Instead, our understanding is that an air bubble from one conduit moves into a neighboring conduit through pits, which is much better described by "embolism propagation" than cavitation.

The change makes sense and was applied to all mentions of cavitation but the one at the end of the discussion where we mention the phase change of water from liquid to gas.

Referee #1 Evaluations:
Recommendation: Minor changes needed

Referee #1 (Comments for the Author (Required)):

In their revised version of this manuscript, the authors have improved the story substantially and I find it clearer and easier to read. The authors have addressed all of my previous concerns and I thank them for their effort on this. Upon reading the new version, I had a few new questions and suggestions (below) but did not have any major concerns or suggestions.

49: The term "disturbance" is vague and I suggest removing it.

Agreed and removed. L:49

65-66: I would argue there is substantial scrutiny about methods in plant hydraulics. There is a whole debate about whether microCT in intact plants provides a better picture than hydraulic measurements on excised segments. I suggest having a look at papers from Andrew McElrone and Anna Jacobsen & Brandon Pratt. Much debate centers around vulnerability but still there is debate about simply measuring conductivity.

The debate does indeed surround the vulnerability of plants and that cutting creates artifacts compared to leaving them intact using microCT. However, microCT establishes a theoretical hydraulic conductivity and not a measured one. We have attempted to emphasize our focus on the hydraulic conductivity measurement methods as follows:
“Many methodological advances have been made to measure the “percent loss of hydraulic conductivity" (PLC) under water stress, but the methods to measure exact hydraulic conductivity values in plants or plant segments have received relatively little scrutiny (Cochard 2013, Venturas

2017). While there is currently a method to measure plant vulnerability in vivo, micro-CT, it does not measure an absolute hydraulic conductivity but a theoretical one (Pratt et al. 2020). Therefore, we will not expand on its details in this paper."
L:63-70

112: This is now very clear why controlling flow rate vs controlling pressure makes a difference.

Thank you for the confirmation.

170-179: Consider a slight rewording that focuses on the flow path from pump to source. The bypasses seem like necessary but not the most important components that can be mentioned at the end.

The section has been slightly reworded, focusing on the flow path with the sensors, then the bypass of the capillary, followed by the filling of the system.
L:173-186

Eqn 2: Check your formatting

Fixed the issue where the fraction covered the whole equation.

206: Are you saying the voltage was 0? If so please provide units. But moreover I find it surprising that the voltage would be zero at atmospheric pressure.

Thank for pointing this out, the units are now provided.
For clarity, we added a couple of sentences to explain how this specific pressure sensor works before mentioning the 0 Volts: "The pressure sensor measures pressure relative to the atmosphere using two openings, one open to the atmosphere and one attached to the volume of interest. A membrane between the two openings bends with the difference in pressures and delivers a voltage based on the amount of inflection. With no difference (both at atmospheric pressure), no voltage is delivered and 0 V was measured. After filling the system with air at atmospheric pressure (assumed to be 101.3 kPa) and recording the measured sensor voltage (0 V), a known change in volume was applied using the syringe and the corresponding pressure was determined from Eq. 2"
L:212-220

213: You state these dates but then don't really mention dates elsewhere i.e. you don't say when measurements occurred relative to calibrations. If you wish to state the date of calibration then please put it in the context of measurements. It's also curious to see calibrations done two years apart but it's not stated why it would be done at this interval. Lastly, consider stating dates with a non ambiguous format such as "10 Dec. 2020" - Or simple Dec 2020 as the exact day of calibration isn't especially important.

We re-did a calibration to check if the sensors measurements drifted over year. We found that both calibrations were very similar and hence pooled together into one calibration curve. The sentence was re-written as:
"In order to determine if the values of the sensors drifted over time, a second calibration was done two years later."
L:225-226

245: Do you mean that 40% of vessels are under 8 cm long and 30% are 8-16 cm long? Then doesn't this mean that 30% of vessels are longer than 16 cm, which would beg the question of why 10 cm was chosen.

40% is the percentage of samples which had a maximum vessel length of 8cm and not the percentage of vessels, we attempted to clarify this as follows:
“For samples in this paper, 10 cm are removed from each end, as Buchmüller (1986) found that 40% of the dry wood samples of *Fagus sylvatica* had a maximum vessel length of under 8 cm, with an additional 30% of samples having a maximum vessel length between 8 and 16 cm. Any branches along the cut segment are removed under water and sealed with parafilm and/or silicon gel to avoid air entry.”
L:256-261

264: Your equation Ks = k/mu does not work out with the units. At the least, your stated k and mu don't contain kg but you state k/mu contains kg.

Thank you for pointing this out, the ρ was missing in the numerator. This has been corrected for both Ks and Kas.
L:277&279

263-269: I would bet the majority of readers would be familiar with conductivity (kg m s-1 MPa-1), specific conductivity (kg m-1 s-1 MPa-1) and conductance (kg s-1 MPa-1). I can't say I've come across specific conductivity as m2 MPa-1 s-1, so would disagree that "many" authors use it. Maybe that form is used in areas of hydraulics outside of plant hydraulics. If you choose to continue to use permeability then I think it's necessary to relate it to the conductivity that people would be most familiar with. I now see you choose to use K as in Eqn 5, which is a form I think most would be familiar with. Because you are using K, I suggest starting that earlier on and omitting the aforementioned list of different metrics that others use.

We agree and have change “many” to “some”.
However, because there are many different forms that can be reported, we felt it is useful to keep the comparison between them.
L:276

360-368: This info seems better in the methods.

The section was moved and integrated into the methods section for the experiment: “The beaker was elevated to 35.5 cm for 30 minutes, then to 73.5 cm for 30 minutes, then to 104.9 cm for 30 minutes, and then returned to 73.5 cm for another 30 minutes, before returning to 35.5 cm for 30 minutes.

After this set of measurements, the beaker was placed back on the table, and the syringe pump was re-attached. Water was pulled through the sample (in the same flow direction as before) at flow a rate of 10 μL min^{-1} for 30 minutes, then at 20 μL min^{-1} for 30 minutes, then at 30 μL min^{-1} for 30 minutes, then again at 20 μL min^{-1} for 30 minutes, before returning to 10 μL min^{-1} for another 30 minutes.”
L:320-328

The first sentence in that part of the results section was adjusted to: “During the Sperry method (Fig. 4a), conductivity decreased steadily by 66% over the total 2.5 hours.”
L:376

379: If you're going to state that rates of decline differ, it would be good to quantify that more specifically e.g. to state rate of decline rather than a total change.

The rate varied over time, but we now provided the relative decrease along with the time over which the decline occurred. (i.e. 66% in 2.5 hours and 50% in 2.5 hours)
L:376-377 & 385

391: It would help to state the ranges where this reference condition was used to make it clearer what "in between" means. The caption suggests d-e is back to the reference condition, but it's not clear if f-g is also.

Agreed, the text was changed to: “This reference scenario was established in Segments `a-b', `d-e', and `f-g', for direct comparison to the stress conditions.”
L:397-399

And the caption of Fig. 5 to: “At `f', the system returned to the same conditions as `a' and `d'.”

411: fix "NPa"

Done
L:418

452: This is the first and only time you call it a "needle"

Thank you for pointing it out, it has been changed to leaf as in the rest of the paper.
L:459

549: There could be many reasons why conductivity would differ between studies including, as you suggest, the difference between using stems with emboli or not, as well as other reasons. So I wouldn't think it's necessary to compare.

Given that the values in conductivity can vary by several orders of magnitude at the intra- and inter-species levels, we found it noteworthy to point out the narrow range of variation we found for this species and in relation to some literature values obtained using other methods for the same species.

556: Technically yes, refilling conduits would increasing the area of xylem that is conducting water, but you define the area as the xylem cross-sectional area, regardless of its functional status.

The statement could be misleading and thus we removed the last part of the sentence.
L:561

561: KCl will change the pH little if at all. KCl is used to try to recreate the ionic properties of xylem sap. Have a look at "More than just a vulnerable pipeline: Xylem physiology in the light of ion-mediated regulation of plant water transport" by Nardini et al JXB, for example. And you might consider the possibility that perfusing the stem with distilled water is gradually affecting pit properties and decreasing conductivity.

Thank you for referring us to this paper. All three of the solutions prevent a decline in conductivity over time in the experiments. Only two of the mentioned substances reduce the pH, HCl or oxalic acid. We attempted to clarify this and added the citation for Nardini et al. as follows: “To avoid the decline in hydraulic conductivity and to mimic xylem sap, it has been suggested to add HCl, KCl, or oxalic acid to the perfusing solution (Sperry et al. 1988, Kolb et al. 1996, Nardini et al. 2011). However, a decline in K under large pressure gradients even with de-gassed KCl solution and bacteriocide was found in a study by Bonetti et al. (2021). This decline could easily be investigated

further using the syringe setup, especially the question whether the decay is due to pressure or flow rate, which cannot be done with any of the current setups."
L:564-571

625: You seem to mostly provide liquid pressures in Pa but sometimes use kPa. Consistency is preferred.

Pressures are now stated in kPa throughout.

Supplement:

Fig S4: Capacitance is a metric with units of e.g. kg MPa-1 m-3. So the red line in the figure is *due to* capacitance but it is not a *measure of* capacitance. I suggest calling it cumulative uptake or discharge (depending on your sign convention) or, as you've labeled the axis, "change in volume" rather than "capacitance".

This is a good point, "capacitance" has been changed to "change in water storage".