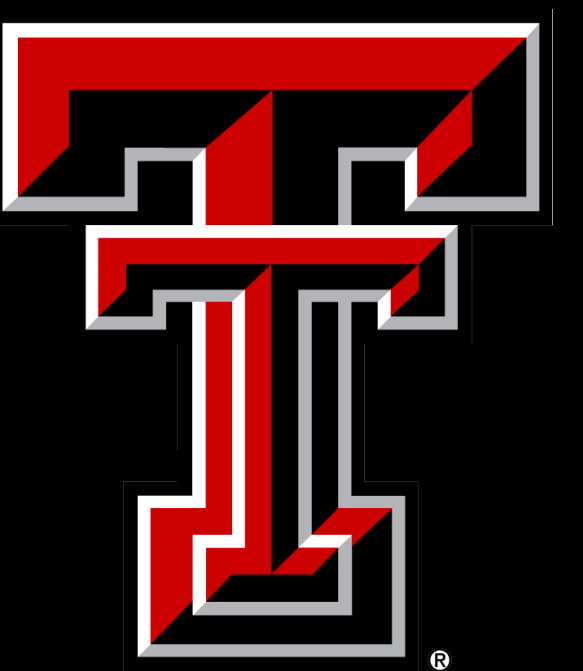




First Report of Use of a Modified Specific Gravity Technique to Determine Viability and Quality of Human Embryos



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Objective

Embryo cryopreservation has been a popular method for preserving fertility as it provides many benefits such as genetic preservation and the easy ability to transport and transfer embryos. This process continues to be carried out in IVF procedures and has led to many successful pregnancies. Although the freezing of embryos may produce many benefits, this method can also have a negative effect on the viability of embryos. Previous studies have demonstrated a modified specific gravity technique useful in determining the health and viability of a number of animal species. The current study is the first report of the use of this buoyancy technique with human embryos. Embryo cryopreservation at the blastocyst stage were thawed, and their buoyancy as it related to their viability, was used to determine the healthiest embryos for use in IVF procedures.

Methods and Material

To thaw the cryopreserved donor embryos, instructions for the Global blastocyst fast freeze thawing kit were used as reference. Human embryos frozen at the blastocyst stage were thawed using the Irvine Fast Thaw kit. Prior to measuring the drop time of the blastocysts, culture plates containing blastocyst culture media were prepared. Also prior to thawing the embryo, a culture plate was prepared with the five thawing solutions and overlaid with mineral oil. After obtaining the embryo straw, the embryo straw was removed from liquid nitrogen and moved to 30 °C water bath. The embryo straw was cut open and the embryo placed into solution one for three minutes. The blastocyst was then moved in sequence through solutions two through five for five minutes per solution.

After the blastocyst was thawed, the individual blastocyst was passed through the Modified Specific Gravity Device (MSGD) which was filled with Global embryo culture media, the same media the embryos were originally cultured in. Prior to dropping the blastocyst, it was rinsed in the first thaw solution to remove mineral oil residue. The amount of time the blastocyst took in passing a one-centimeter vertical distance was measured in seconds. The blastocyst was then recovered, and the diameter of the inner cell mass was measured in micrometers. The blastocysts were also categorized based on hatching and expansion. After incubating in culture medium, the blastocyst was dropped a second time at 6 hours. The blastocysts then were placed back in the 37°C incubator to watch for hatching by 24 hours post-thaw.

Comparisons between descent times were made between groups of embryos that expanded and those that did not, and those that hatch and did not using independent Student T-tests.

Results

- No significant difference in average diameter at thaw across all blastocysts
- No significant difference between initial drop times for expanded vs. non-expanded blastocysts
- Expanded blastocysts had a longer secondary drop time as compared to non-expanded ($P < 0.04$)
- Hatched blastocysts had lower initial and secondary drop times compared to non-hatched ($P < 0.001$ and $P < 0.05$ respectively)
- Differences between initial and secondary drop times were similar for both hatched vs. non-hatched blastocysts

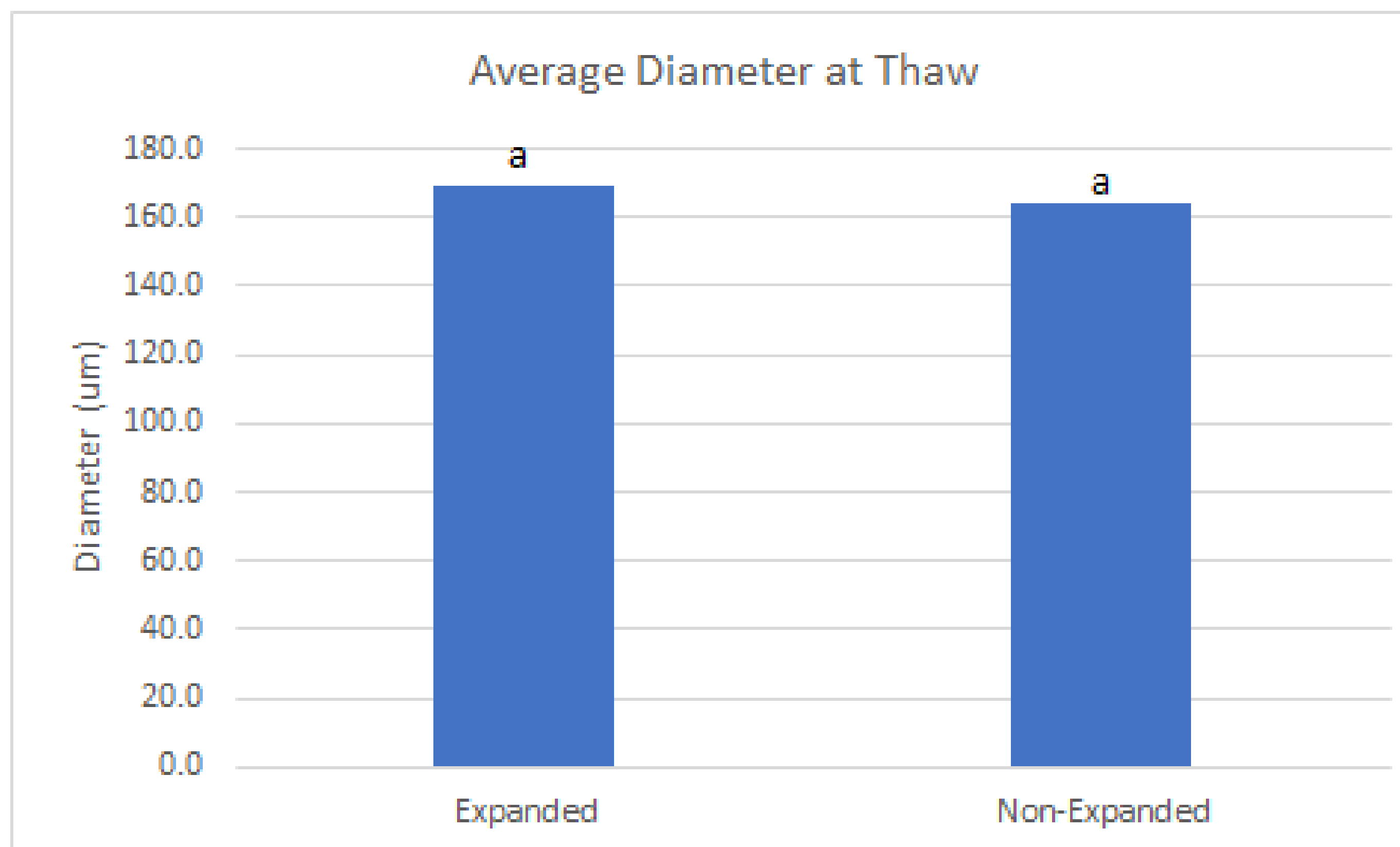


Figure 1. Average diameter of expanded and non-expanded blastocysts after being thawed.

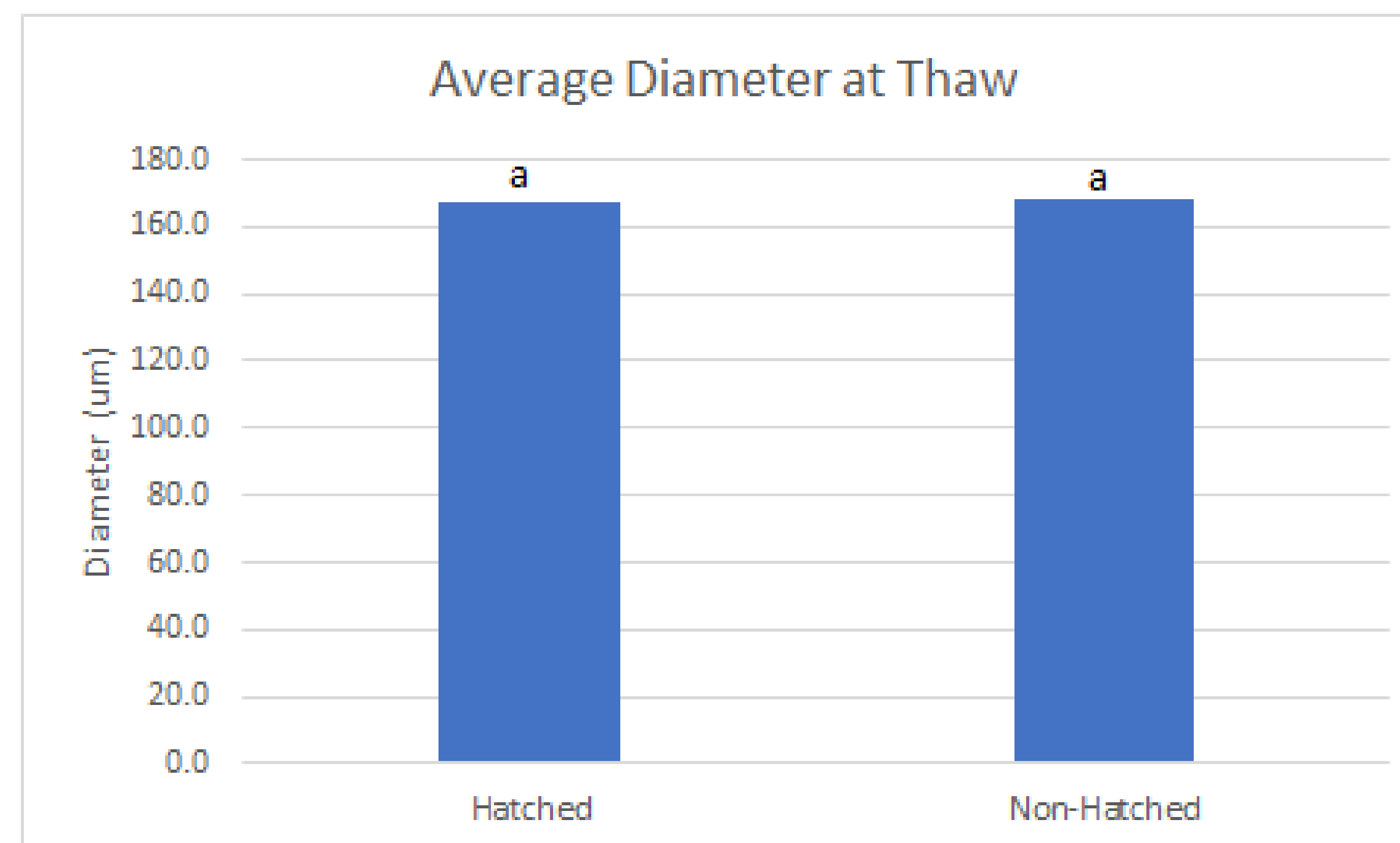


Figure 2. Average diameter of hatched and non-hatched blastocysts after being thawed..

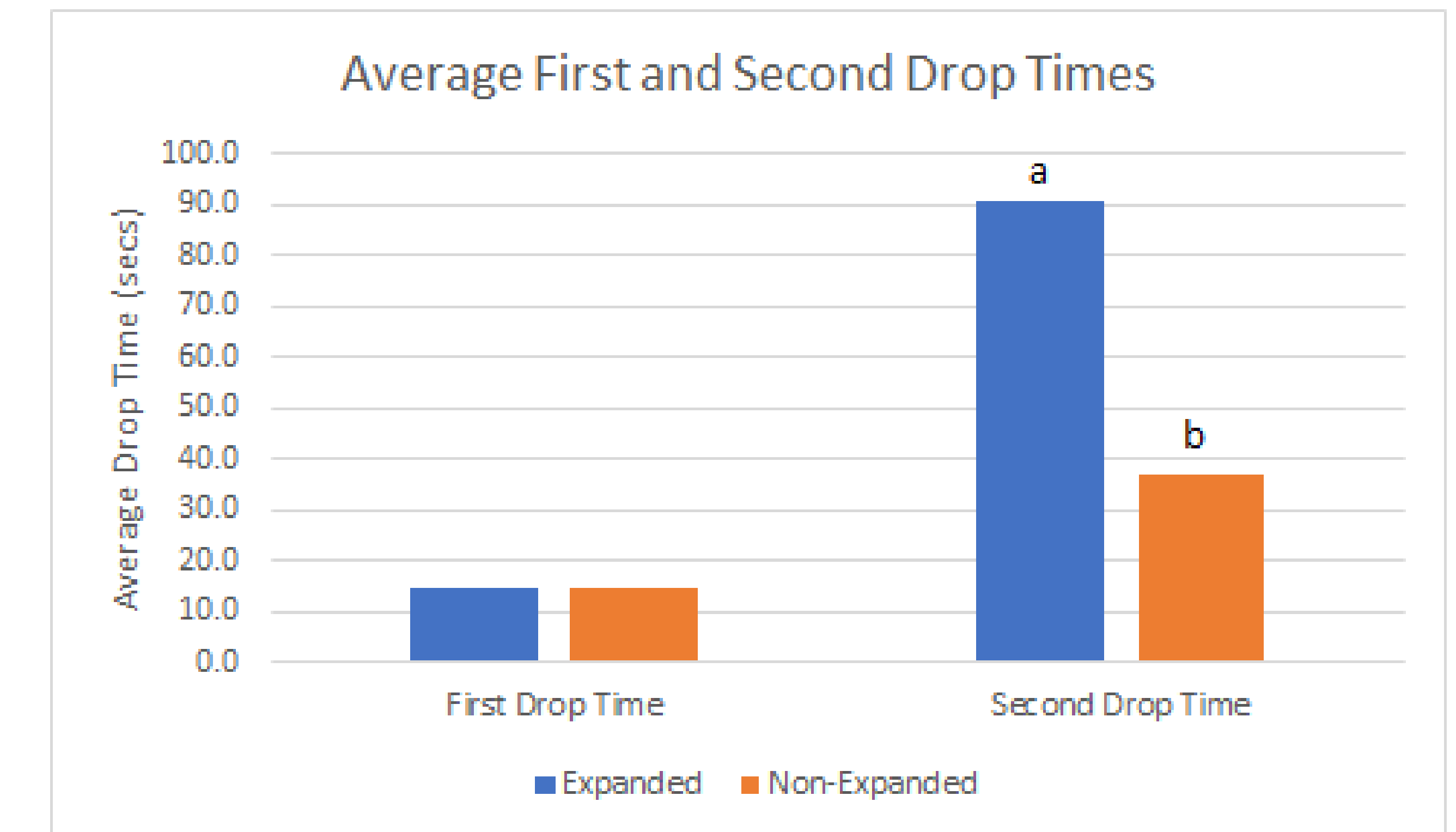


Figure 3. Average drop times of expanded and non-expanded blastocysts.

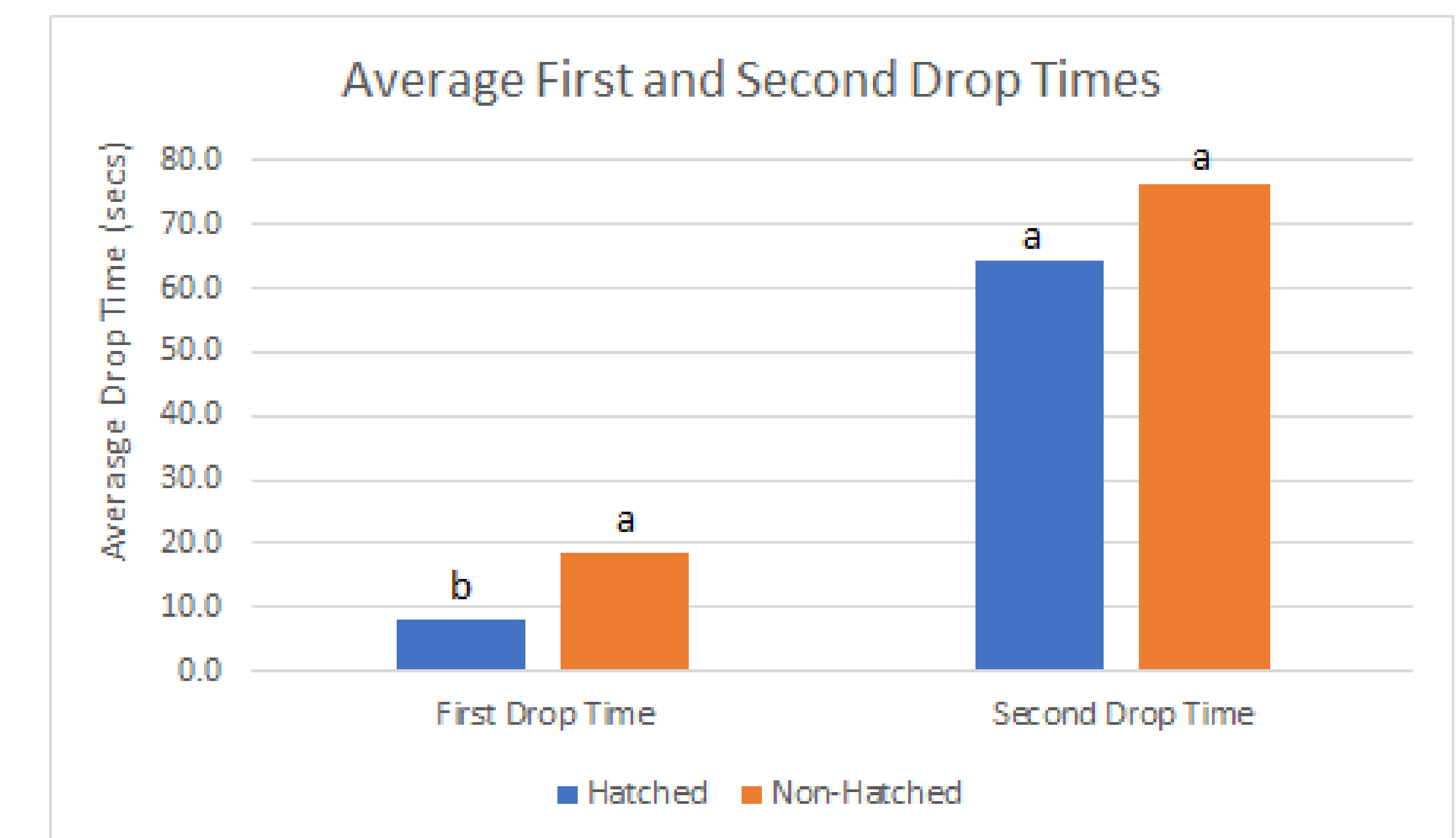


Figure 4. Average drop times of hatched and non-hatched blastocysts.

Conclusions

Preliminary results of these first studies with human embryos demonstrated potential differences in descent times among expanded and hatched blastocysts before and after incubation. Further studies are needed to evaluate this assessment technique on a larger scale. Overall, the data suggests buoyancy may be used as an indicator for embryo viability, increasing conceptions rates through IVF.

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