

O-137 The impact of dry incubation on osmolality of media in time-lapse culture dishes

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Study question: Does continuous culture in a dry incubator lead to an increase in culture medium osmolality that is affected by the type of time-lapse dish used?

Summary answer: Osmolality increased for all tested time-lapse dishes, with a greater effect for Primovision, Miri-TL and standard microdrop culture compared to Embryoscope dishes.

What is known already: Osmolality of culture media is critical for optimal embryo development with high osmolality having adverse effects. Osmolality of culture media is affected by factors such as humidity, temperature, dish preparation method and media droplet volume. Compared to humidified incubation, negative effects of dry incubation on human embryo development have been observed. However, the use of dry incubators is becoming increasingly common due to advantages of time-lapse continuous culture. Different time-lapse systems use different dishes with various media to oil surface ratios, which may alter the effect of dry incubation on osmolality.

Study design, size, duration: Changes in osmolality were measured in four culture dishes over 7 days in a dry incubator (Miri-TL). The culture dishes were: Miri-TL culture coin (MTL), Vitrolife EmbryoSlide (ES), and Primovision dish (PV), with a Vitrolife Micro-Droplet dish (MD) as the control. The study was repeated three times. Osmolality was determined in duplicate at days 0, 3, 5 and 7 of culture. The impact of oil depth on osmolality was also studied for standard MD dishes.

Participants/materials, setting, methods: Dishes were prepared with protein supplemented media and oil. For MD dishes, the impact of 3, 5 or 7 mL oil overlay was determined over 5 days. The time-lapse dishes were prepared as per manufacturer. MD dishes had 20 μ L microdroplets under 5 mL of oil. For each condition, osmolality was tested on a 10 μ L sample using a VAPRO osmometer. Relative changes in osmolality were assessed (ANOVA) and means were compared using Tukey's HSD.

Main results and the role of chance: Culture media osmolality was affected by oil depth, with a 3 mL oil overlay resulting in the highest osmolality compared to either 5 or 7 mL. For time-lapse dishes, osmolality increased as the culture continued over 7 days in the dry incubator. From a starting osmolality of 258 mOsm, media osmolality in MD, MTL and PV increased similarly over 7 days of culture, but for ES dishes, the increase in osmolality occurred to a lesser extent, showing that the ES condition was less prone to increasing osmolality during a 7 day dry incubation. For MD, MTL, PV and ES dishes, osmolality increased, respectively, by 26.9, 23.7, 20.3 and 12.2 mOsm on day 3, 42.3, 42.5, 38.5 and 27.7 mOsm on day 5, and 53.5, 50.4, 57.3 and 33.2 mOsm on day 7. ES osmolality was lower than the control MD dish on day 3 ($p < 0.05$), both MD and MTL on day 5 ($p < 0.05$) and all dishes on day 7 ($p < 0.01$), showing that the ES condition had the greatest ability to prevent media evaporation, resulting in slower osmolality rise over 7 days of culture during dry incubation.

Limitations, reasons for caution: This study was performed using a MiriTL bench top incubator; therefore the results may not be applicable to other dry incubators. Osmolality increased in all dishes and the impact of these increases on human embryo development is unknown.

Wider implications of the findings: The osmolality of culture media increased with dry incubation despite oil overlay in various dishes, an increase that was dish dependent. The ES dish, which uses deep culture wells that results in the lowest ratio of culture medium surface area to oil, had the lowest amount of media evaporation.

Trial registration number: Not applicable.