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Set-up of a ^{t}WE basic ART laboratory

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INTRODUCTION:
Parallel increases in Assisted Reproduction Technology (ART) regulations and specifications as well as growth in the commercial ART industry has occurred in the last decade, with a concurrent escalation in cost. The question arises what is the suitability and requirements of an ART procedure for its intended purpose or outcome? Deviations outside of the physico-chemical boundaries of culture media/conditions, i.e. osmolality, pH, temperature will impact negatively on clinical outcome. The Walking Egg (a non-profit organization), designed a ^{t}WE lab ART method specific for low resource settings, whereby medical gases, complex incubation equipment and infrastructure are not required.

AIM:
The aim of this study was to investigate some of the physico-chemical parameters of the ^{t}WE lab ART method, i.e. temperature and pH stability during culture.

MATERIALS AND METHODS:
Experiment 1: Stoppered glass tubes with 1ml culture medium (LifeGlobal® Global® Total® for Fertilization) in aluminium blocks (used in the ^{t}WE project) using 3 different heating devises generally used in an ART Laboratory set at set-point 37°C, i.e. H1: *closed* - a warming oven (Quincy Lab Model 10-140E Incubator), H2: *partially closed* - a dry bath (K-Systems Dry bath DB-006), used for the heating of aspiration tubes and H3: *a single surface heater* – a slide warmer (Adamas Slide Warmer SW85). The temperatures of the culture media were monitored *in situ* using a calibrated three decimal electronic thermometer (Greisinger Digital Thermometer GMH G230) equipped with a wire probe.

Experiment 2: The heating device which provided optimal results was selected and incubation of the culture media with different CO_2 gas volumes was simulated. CO_2 was generated by adding citric acid (CA) to bicarbonate of soda (BoS) in stoppered glass tubes. Sibling glass tubes with CO_2 being generated was connected via tubing to culture media containing tubes. Ten volumes of CA (1.2ml – 3.0ml at 0.2ml increments) were added to an excess amount of BoS. The tubes were incubated at 37°C for 18 hours and the pH of the culture media was measured with a blood gas analyser (Radiometer ABL 800 Flex).

RESULTS:
Positioning of a warming block with the test tubes impacted on temperature readings. Five tubes per block with 10 repeats indicated the following temperature measurements:
H1: 37.2±0.28°C, 36.2±0.38°C (left & right side of device); H2: 36.7±0.18°C (for both left & middle position); H3: 36.2±0.16°C and 36.7±0.09°C (front & back position) (CV 0.74%, 1%, 0.46%, 0.47%, 0.23%, 0.43%).

Culture media pH after gassing with CO_2 produced by increasing volumes of CA (1.2–3.0ml, with 0.2ml increments) was 7.421±0.056, 7.382±0.045, 7.325±0.046, 7.314±0.032, 7.257±0.060, 7.241±0.038, 7.180±0.055, 7.164±0.039, 7.133±0.065, 7.106±0.074 (CV 0.76%, 0.61%, 0.62%, 0.44%, 0.83%, 0.53%, 0.77%, 0.55%, 0.91%, 1.04%).

CONCLUSION/DISCUSSION:
Optimal temperature regulation was achieved with using the dry bath with blocks, which is a cost-efficient device. The fluctuation in temperature of the warming oven may be due to indirect heating through air, while direct heat transfer can be controlled more accurately using a slide warmer or dry bath. Temperature variances per temperature regulating device occurs more frequently than anticipated in the ART Laboratory.

As anticipated, gassing of culture media with increasing volumes of CO_2 provides a linear decline in pH. The choice of media and location of the laboratory will impact on the setup of the culture system. Practical aspects influenced the pH values of the culture media. The system design was addressed to prevent fluctuations.