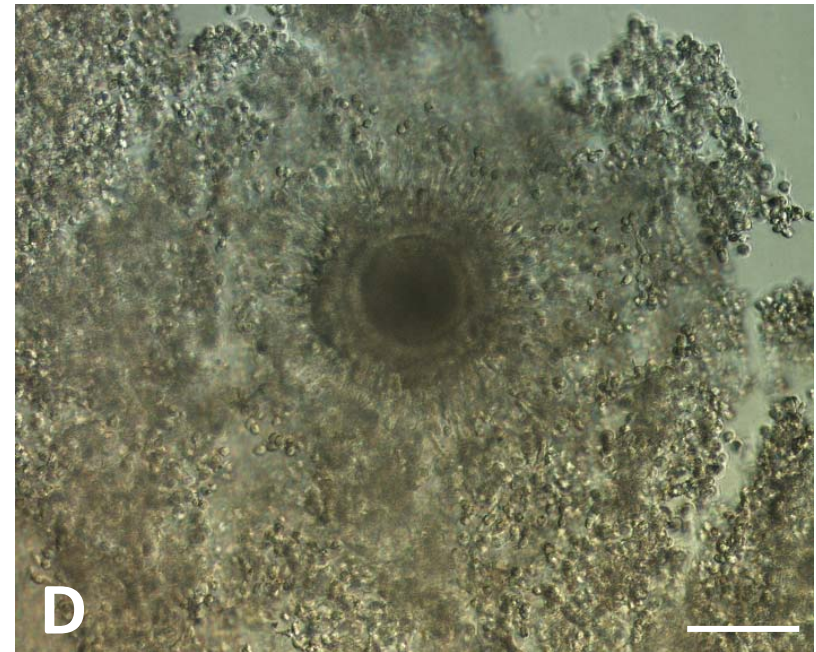
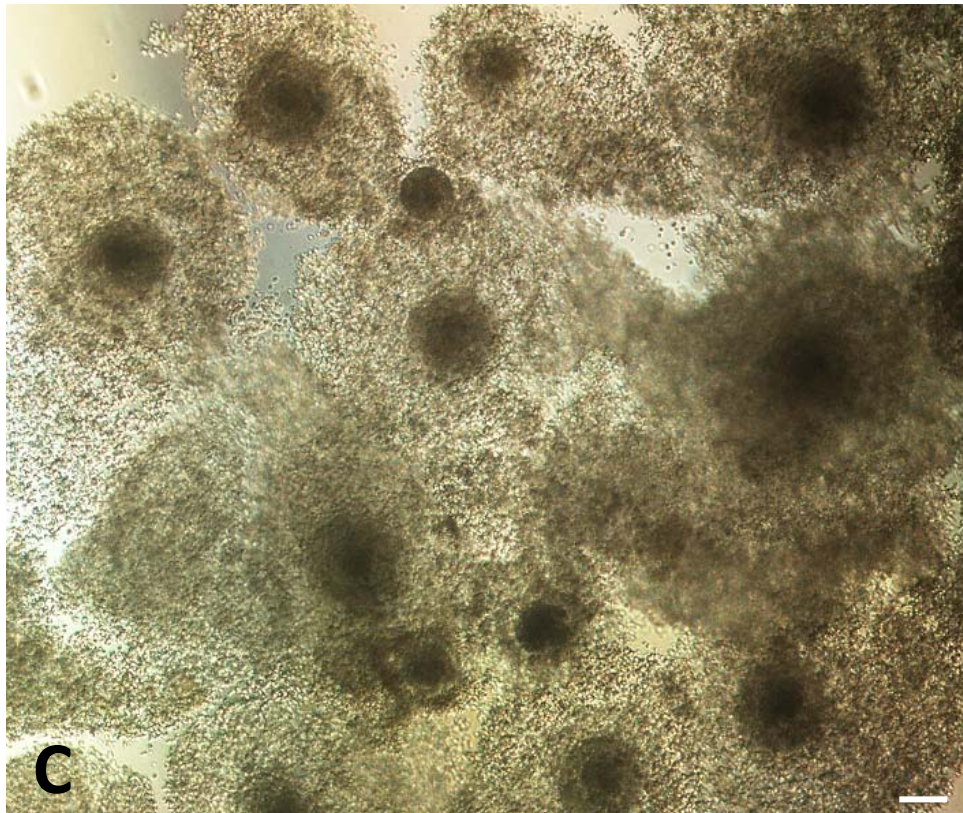
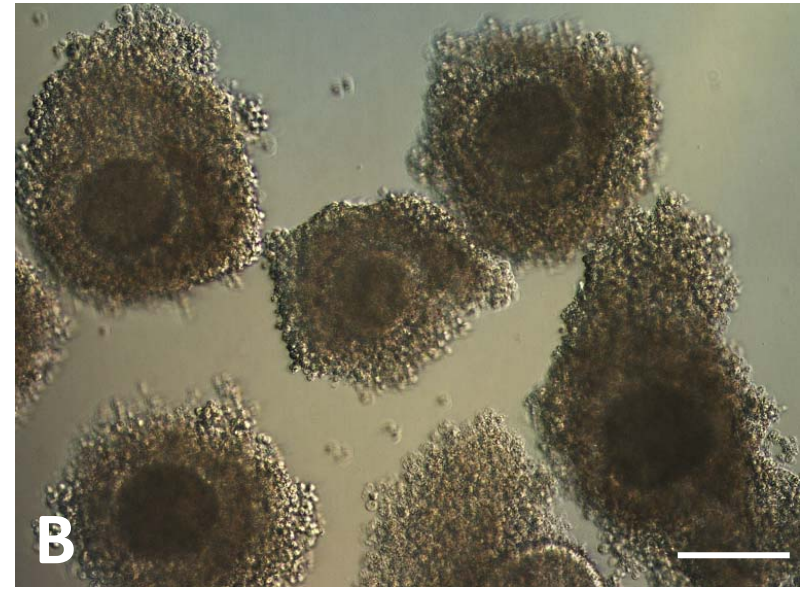
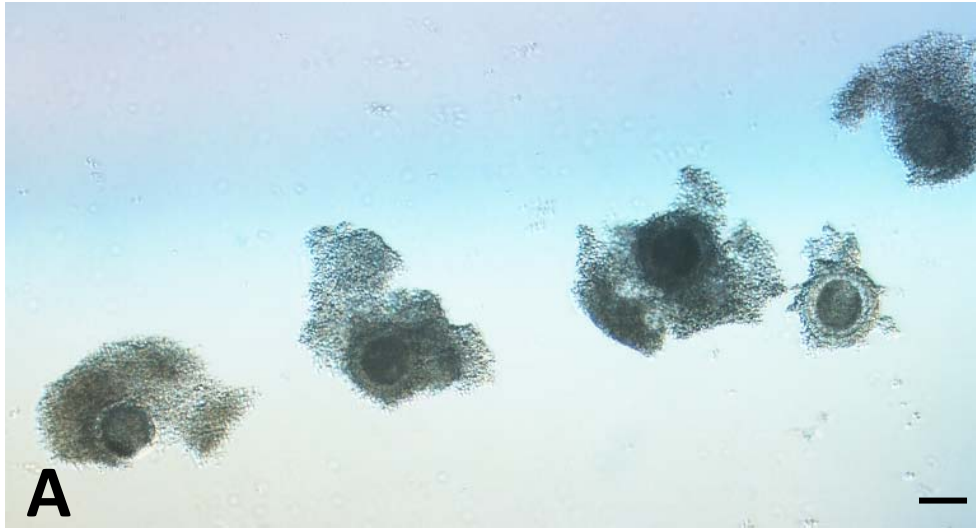


Oocyte maturation and fertilization *in vitro* with RLI's OAP, OWP

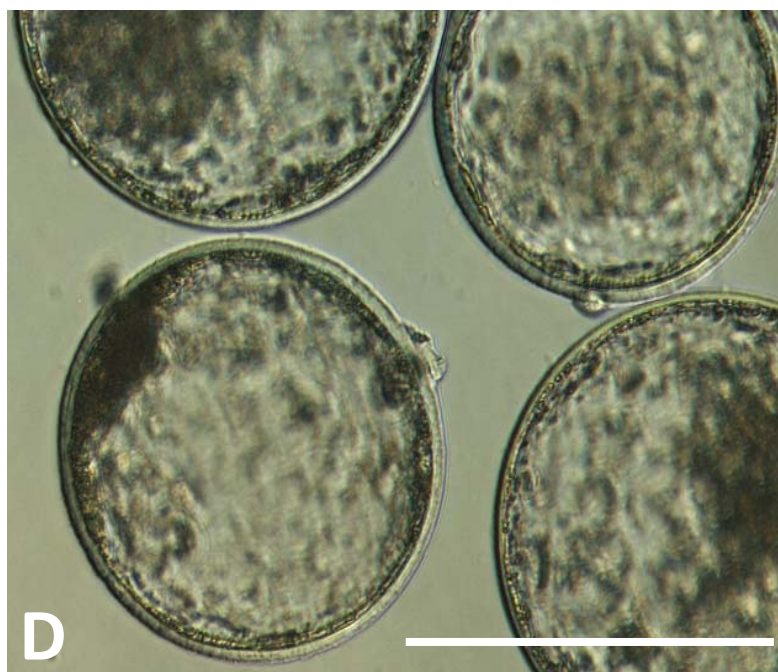
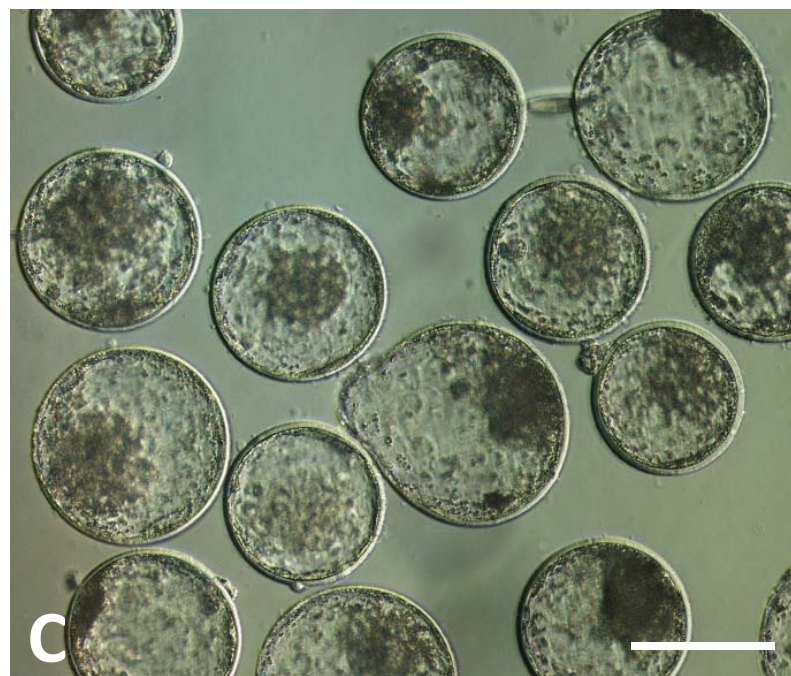
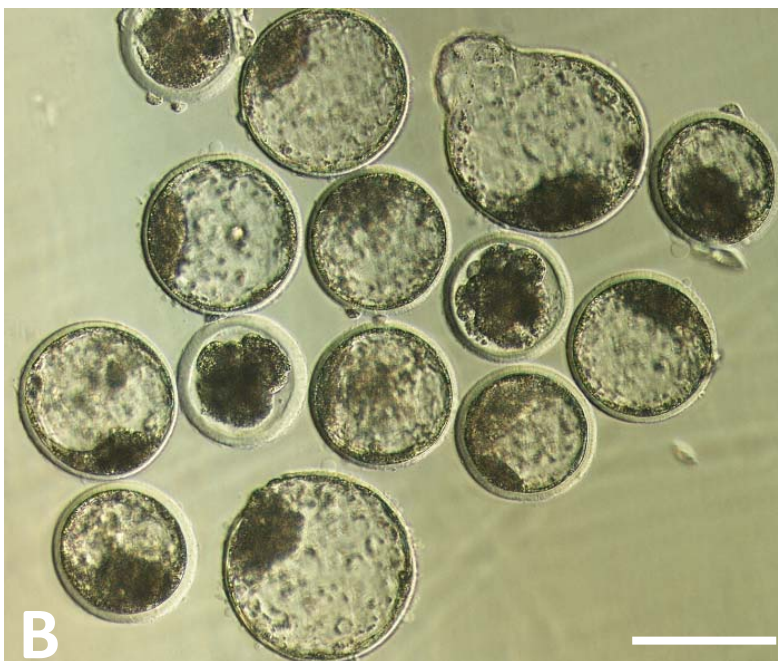
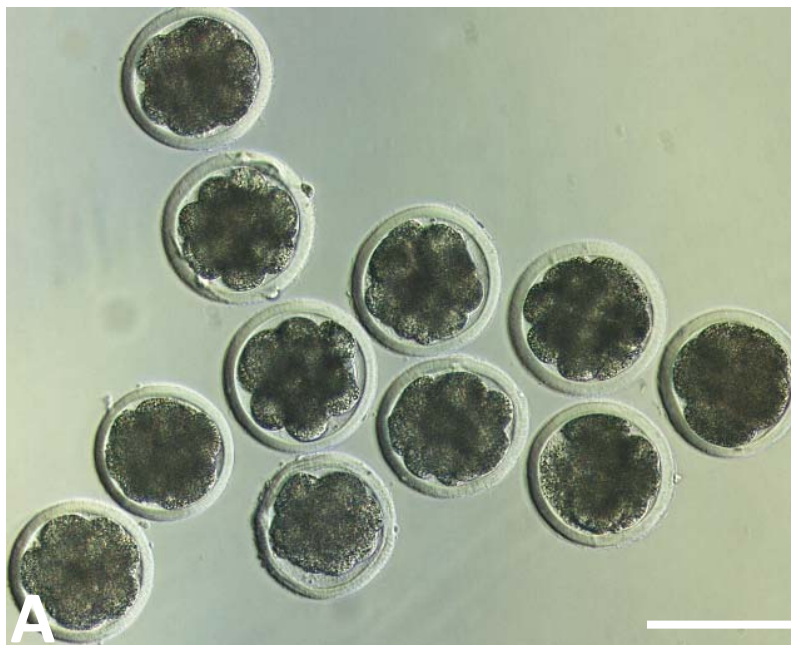
Renova Life Inc.

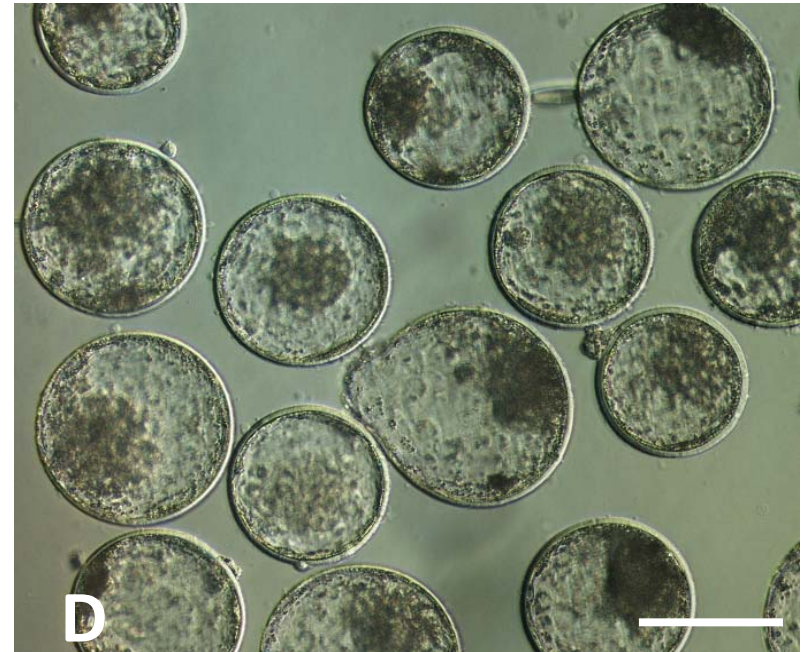
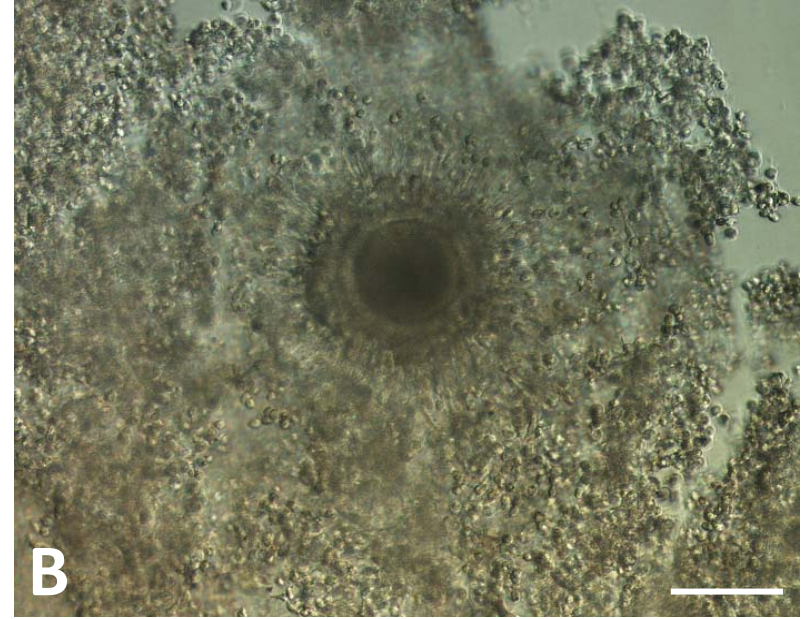
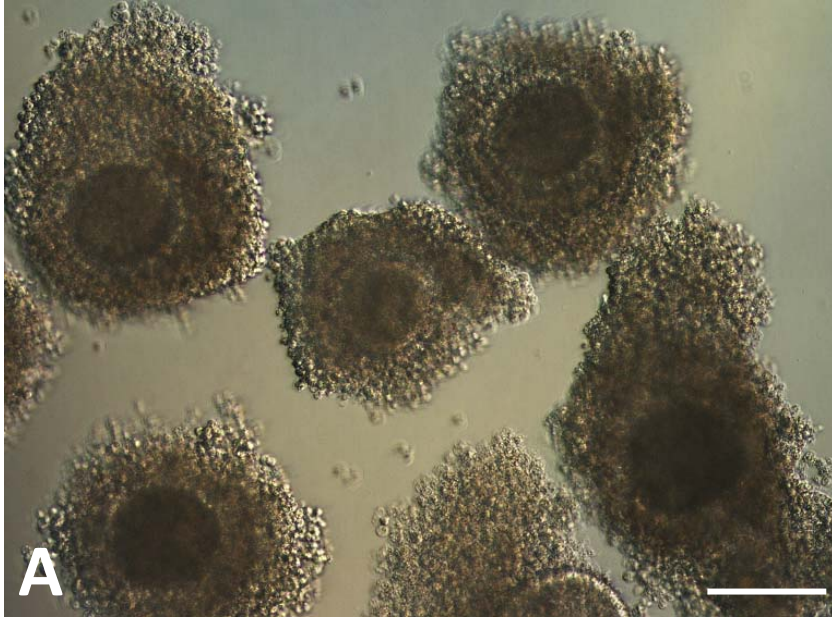
2014

- Oocytes were collected from slaughterhouse ovaries on 01/29/2014. Oocyte-cumulus complexes (OCCs) were aspirated with Renova Life Inc.'s (RLI) oocyte aspiration plus (OAP) , lot # 01172014-023OAP, and washed three times with oocyte washing prior to maturation (OWP) , lot #01292014-005OWP before put into maturation medium.
- RLI sent the same lot batch of OAP and OWP to Maryland office.
- Maturation medium (MAT) was consisted of M199+FCS supplemented with 0.5 µg/ml ovine FSH, 5.0 µg/ml ovine LH (NIDDK) and 1.0 µg/ml estradiol (Sigma, E-8875).
- OCCs were washed in MAT one time, and transferred into 75 µl/drop MAT covered with mineral oil, and cultured at 39°C, humidified 5% CO₂ for 22 h. This was done on 01/29/2014.
- OCCs were taken photos at 0 hr of maturation (01/29/2014) and 22 h after IVM (01/30/2014) (see figure). After maturation, OCCs were well expanded in the dishes.
- IVF was performed with TALP-Fertilization system on 01/30/2014, embryos were cultured in CULT-medium added with 6 mg/mL BSA (RLI Water-embryo, W-embryo 02: 500 mL), at 39 °C , in O₂, CO₂, N₂ mixed gases for 7 days .
- The development to blastocyst was recorded at day 7 after culture. The percentage of 45% total blastocyst and 40% C1 grade embryos was achieved, respectively.



A, 0 hr IVM, 4X; B, 0 hr, 10X; C, 22h, 4X; D, 22h, 10X. Bar=150 μ m





**Table 1. Fertilization and Embryo Development in vitro with RLI's OAP,
OWP**

Treatment	No. oocytes	No. 2-8 cell (%)	No. 8-cell (%)	D7 Blastocysts (%)	C1 Blastocysts (%)
RLI Water-embryo	80	69 (86)	53 (66)	36 (45)	32 (40)