Scientists announce mouse sperm cryopreservation breakthrough

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A team of Jackson Laboratory scientists have figured out a simple, cost-effective process to freeze mouse sperm and get it to achieve high fertilization rates with mouse eggs. The breakthrough will greatly reduce the cost of developing and distributing new mouse models of human disease.

Freezing sperm is an efficient, cost-effective way to conserve and distribute genetics in the agricultural industry and putting male sex cells on ice is a fundamental part of human fertility programs. But the sperm of certain varieties of mice under-achieve woefully after being frozen and thawed. What's worse: the thawed sperm of the most popular mouse strain in the scientific world, the C57BL/6 or "Black 6", are known to under-perform when it comes to fertilizing mouse eggs.

Drs. Michael Wiles and Chuck Ostermeier in Jackson's Technology Evaluation and Development group, and Dr. Robert Taft and Ms. Jane Farley in the Reproductive Sciences group, have published a paper on the new technique in the open-access journal *PLoS ONE*, where it can be freely accessed online.

The technology has already attracted interest from international academic and pharmaceutical laboratories.

The Jackson team reports that their technique consistently yields fertilization rates of about 70 percent – a six-fold increase over previous mouse sperm freezing techniques. The results were achieved by

collecting the sperm into a cocktail of raffinose (a plant-based sugar complex), skim milk and the antioxidant monothioglycerol. The sperm suspension is loaded into narrow plastic straws about the size of a swizzle stick, and then slowly cooled before storage in liquid nitrogen.

When frozen sperm are needed for fertilization, they are thawed and incubated in in vitro fertilization media for an hour before adding oocyte cumulus masses (clusters of egg cells).

Dr. Wiles noted, "The world research community is making literally thousands of new mouse models," using stem cells to introduce specific genetic variations that mimic the mutations identified in human diseases. "The problem is that it costs about $10,000 a year to maintain a particular mouse strain, and worldwide only a few hundred strains are in actual laboratory experiments at any given time."

Since the 1970s, the Laboratory has addressed this problem by cryopreserving – freezing and storing – mouse embryos from little-used strains, which allows the live mice from those strains to be safely removed from the mouse room. However, freezing embryos is far less efficient and cost-effective than freezing sperm. "If you freeze 250 embryos," Dr. Wiles said, "you can only count on about 125 live pups. But a single male mouse can produce millions of sperm, which can give rise to 100s or even 1,000s of offspring. Thus, making sperm cryopreservation work has long been a goal of ours."

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