

aerobic flush preservation that has stood the test of time. However, we in fact have known since then, that 10°C is in fact a superior temperature for storage of lungs. We have recently shown that a combination of 10°C storage and normothermic EVLP can in fact protect and preserve lungs for up to 3 days followed by successful transplant in a large animal transplant model. This will not only enable an increase in the number of lungs transplanted, but also will make transplantation a more attractive career for future generations of surgeons. The science of transplantation continues to evolve and novel practices that are being translated to the bedside are radically changing the opportunities to save lives of patients with end stage organ failure with improved short- and long-term outcomes.

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CRYOBIOLOGY 113 (2023) 104595 104599 CELLULAR THERAPY AND THE CRITICAL ROLE OF BIOPRESERVATION

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Successful biopreservation is critical to ensure the highest quality of cellular therapies. This presentation will provide an overview of the field of cellular therapy with specific examples highlighting the impact of biopreservation and the need for continued research and optimization.

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CRYOBIOLOGY 113 (2023) 104595 104600 FERTILITY PRESERVATION USING OVARIAN TISSUE FOR WOMEN AND RHINOCEROS

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Fertility preservation by cryopreserving ovarian tissue is required for many prepubertal patient groups, but is also an important method for endangered species. The Northern White rhino is functionally extinct with just two females left. To preserve this iconic species, numerous advanced assisted reproductive technologies and strategies have been utilised around the world. The strategy chosen for the Rhino Fertility Project is to harness the potential of the ovarian primordial follicle reserve by culturing ovarian tissue with the aim of generating eggs in vitro; a process that has been carried out for model species. Through working on the rhino tissues, we have gained insights, identified challenges, and developed new methods that also benefit our work with human ovarian tissues. Obtaining rhino ovaries to develop such techniques has also proved challenging and this void contributed to the creation of Nature's SAFE (Saving Animals From Extinction); a UK biobank charity.

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CRYOBIOLOGY 113 (2023) 104595 104601 CHALLENGES IN DEVELOPING CRYOPRESERVED PfSPZ VACCINES AGAINST MALARIA

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Throughout recorded history, malaria has exacted a huge toll in mortality, morbidity, and disability adjusted life years; currently, annual infections exceed 247 million with 619,000 deaths. Essential for eradicating infectious diseases (e.g. smallpox, polio) is a highly effective vaccine: our goal is a vaccine with >90% protective efficacy. PfSPZ vaccines are cryopreserved: they target *Plasmodium falciparum* (Pf) malaria, and employ metabolically-active sporozoites (SPZ) as the immunogen. PfSPZ are grown, from Pf gametocytes initiated from cryopreserved Pf erythrocytic stage cell banks, in aseptically-reared *Anopheles stephensi* mosquitoes, while cryopreserved eggs provide insurance against mosquito colony collapse. PfSPZ vaccines are attenuated by irradiation, co-administration of an anti-malaria drug or genetically but are all manufactured by an identical process. Specific challenges have included developing a completely aseptic production process, fill-finish and cryopreservation adapted to the time constraints imposed by cryoprotectant additives, development of a new cryovial with septum that maintains container closure integrity below -150 °C and compatible with high throughput, automated thawing, and use at clinics in remote locations, and vaccine administration route, dose and interval. A cold chain employing liquid nitrogen vapor phase cryoshippers, misperceived as a problem by the conventional vaccine industry, is being aided by the advent of CAR-T and cellular therapies. Our cryoshippers are transported by select couriers and carried the last mile by our personnel. They remain, with weekly LN2 recharging, on site as local storage units, sometimes for several months while vaccine is administered. To date, 38 clinical trials at 19 sites in the US, Europe, sub-Saharan Africa and Indonesia, have demonstrated unprecedented tolerance, and achieved up to 100% protective efficacy. Reverse cryologistics have returned unused vaccine, peripheral blood mononuclear cells and other samples.

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CRYOBIOLOGY 113 (2023) 104595 104602 THE DEVIL'S ADVOCATE REGARDING USE OF ME₂SO FOR CRYOPRESERVATION OF CELL AND TISSUE TRANSPLANT THERAPIES

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Dimethyl sulfoxide (Me₂SO) is a commonly used cryoprotectant in cell and tissue cryopreservation, but its application is not without drawbacks. While Me₂SO has several beneficial properties that aid in preserving cells and tissues, it also presents challenges and potential adverse effects for clinical cell and tissue therapies. The Good: Me₂SO is an effective cryoprotectant due to its ability to penetrate cell membranes, replacing intracellular water, reducing intracellular ice formation, and preventing cell damage during freezing and thawing processes. Me₂SO has been shown to be compatible with many cell and tissue types without which most clinical cryopreserved cell therapies would not be possible. The Bad: Me₂SO has inherent cytotoxic properties, which can cause damage to cells and tissues. The concentration, temperature, and duration of Me₂SO exposure must be carefully controlled to minimize toxicity. Me₂SO can induce cellular stress responses, affecting cell behavior, metabolism, and gene expression patterns. Me₂SO has been shown to interfere with cell differentiation processes, potentially altering the phenotype and functionality of cryopreserved cells, chromosome damage and mutations. The Ugly: There is an extensive literature on clinical side effects of Me₂SO. Frequently observed acute effects include skin irritation, diarrhea, vomiting, odor, and taste disturbances. Me₂SO is classified as a drug, and the US Food and Drug Administration (FDA) has attempted to ban Me₂SO on more than one occasion, but there is presently no proven replacement. Reports of extreme changes in microRNAs and alterations in the epigenetic landscape suggest that Me₂SO is not inert. Such reports have led to the suggestion that the use of Me₂SO should be reconsidered, especially for cryopreservation of embryos and oocytes, since it may impact embryonic development. In conclusion, Me₂SO offers significant benefits as a cryoprotectant for cell and tissue preservation. However, it is essential to