Journal of Equine Veterinary Science 43 (2016) S29–S36

Contents lists available at ScienceDirect

Journal of Equine Veterinary Science

journal homepage: www.j-evs.com

Review Article

Recent Developments in Stallion Semen Preservation

Zamira Gibb*, Robert J. Aitken

Priority Research Centre for Reproductive Science, Discipline of Biological Sciences, Faculty of Science and IT, University of Newcastle, New South Wales 2308, Australia

ARTICLE INFO

Article history:
Received 15 April 2016
Received in revised form 8 June 2016
Accepted 8 June 2016
Available online 18 June 2016

Keywords:
Stallion
Semen preservation
Spermatozoa
ART
Artificial insemination

ABSTRACT

Since the commercialization of artificial insemination in the early 20th century, there has been a need reduce the logistical difficulties associated with the coordination of semen collection and insemination. This need has been met through the development of various approaches to extend the longevity of spermatozoa through semen dilution and temperature-induced metabolic restriction, and these strategies have provided livestock breeders with a valuable tool for the genetic improvement of many species. As the horse is not a production animal, the resources available for the research of stallion semen preservation have been scarce. Despite this, significant progress in semen processing and preservation techniques has been made in the areas of chilled storage, cryopreservation, and more recently ambient temperature storage. While the major focus for research in the areas of chilled and cryopreserved spermatozoa has been the stabilization of membranes during the deleterious phase changes that occur throughout cooling, ambient temperature storage presents a completely different set of problems associated with the rapid metabolic rate of stallion spermatozoa which use oxidative phosphorylation and produce vast quantities of reactive oxygen species. This article reviews the tactics that have been used to overcome the damaging effects of semen preservation which are unique to each strategy, while touching on the fundamental discoveries which have brought the field to the position it is in today.

© 2016 Elsevier Inc. All rights reserved.

1. Introduction

Horses are selected for breeding on the basis of pedigree and athletic performance as opposed to reproductive traits and for that reason are not subjected to selection pressure for fertility. Fertility traits are heritable [1], and the practice of circumventing subfertility through the use of assisted reproductive technologies (ARTs), because it places no importance on reproductive fitness in the selection of breeding animals, has resulted in equine populations with significantly lower per-cycle conception rates than other species [2]. Artificial insemination (AI) is a widely used tool in modern horse reproduction [3], with around 90% of Standardbred foals being produced via AI of chilled or cryopreserved stallion spermatozoa [2]. For its part, the use of AI brings a number of advantages, such as the prevention and control of disease through the eradication of direct male-to-female contact, an increased rate of genetic gain through the importation of new genetics, and the preservation of spermatozoa for later use in case of death or infertility.

The storage of stallion spermatozoa is necessary for a number of reasons associated with ART such as AI and intracytoplasmic sperm injection (ICSI). During the final phases of spermatogenesis, spermatozoa lose the ability to biosynthesize and repair and consequently become extraordinarily simple in their metabolic functions [4]. The inevitable cell demise that follows can be delayed or even prevented through the application of temperature-induced metabolic restriction by chilling or cryopreservation, depending on the required duration of storage. If AI is to be

* Corresponding author at: Zamira Gibb, Discipline of Biological Sciences, Faculty of Science and IT, University of Newcastle, Callaghan, New South Wales 2308, Australia.
E-mail address: zamira.gibb@newcastle.edu.au (Z. Gibb).

0737-0806/$ – see front matter © 2016 Elsevier Inc. All rights reserved.
http://dx.doi.org/10.1016/j.jevs.2016.06.006

performed within hours, spermatozoa are generally left at ambient temperatures. If sperm longevity must be maintained for longer periods, spermatozoa are either chilled (up to 72 hours) or cryopreserved (indefinite) to restrict the metabolic rate following dilution in an appropriate semen extender. Rumor has it that the horse was the first species in which semen was diluted for the purpose of AI. In 1322, the semen from a much admired Arabian stallion was stolen from the vagina of a recently bred mare, diluted with camel's milk, and used to inseminate the mare of a neighboring rival chieftain with the reported birth of a foal the following year [5]. Many years later, the phenomenon of cold-induced sperm preservation was first discovered by Spallanzani [6] in 1776. He observed that cooling of frog, stallion, and human semen in snow did not kill all the "spermatic vermicules," but rendered them temporarily immotile and induced a state of lethargy from which they could recover when returned to higher temperatures. By restricting the metabolic rate of cells, the production of toxic metabolic by-products, such as hydrogen peroxide, lipid aldehydes, and carbon dioxide, are reduced and the depletion of adenosine triphosphate (ATP) associated with the maintenance of homeostasis [4,7] is minimized. This temperature-induced metabolic restriction reduces the rates of both reactive oxygen species (ROS) production and acidification of the storage medium through the accumulation of lactic acid and CO2 from glycolysis and OXPHOS, respectively. However, the spermatozoa of many stallions do not tolerate the stressors associated with chilling or cryopreservation particularly well [8,9], and following on from studies investigating ambient temperature semen storage using extenders intended for chilled semen [10], there has recently been a concerted effort to develop a synthetic medium which will extend the longevity of stallion spermatozoa without the need to chill or cryopreserve [11].

This review assembles recent developments in stallion semen preservation technologies to summarize the currently accepted "best practice" protocols in chilled, cryopreserved, and ambient temperature semen storage, while commenting on the novel and emerging methodologies that are on the horizon.

2. Developments in Semen Processing for Storage

To remove seminal plasma or not to remove seminal plasma? That is the question. While the presence of seminal plasma has been shown to be beneficial to the viability and acrosome integrity of spermatozoa from boars, bulls, and rams [12], it tends to be detrimental to the motility and membrane integrity of stallion spermatozoa, and its removal by centrifugation is almost always advocated [13-15]. Despite this, several investigators have found the presence of a small amount of stallion seminal plasma (0.6%–20%) to be beneficial to both sperm parameters [16,17] and fertility, the latter possibly being due to its inhibitory effect on the binding of polymorphonuclear neutrophils, reducing sperm phagocytosis within the female reproductive tract, and enhancing fertility when there is an underlying uterine inflammation [18]. There is clear evidence to suggest that the inclusion of very small amounts of seminal plasma may be beneficial or at worst have no deleterious effects. Unless a density gradient or multiple washes are used to isolate sperm cells before processing then there will always be a small amount of seminal plasma present in a centrifuged, resuspended sample, and therefore, in the majority of clinical situations, seminal plasma replacement ought not to be necessary.

The majority of protocols for the isolation of spermatozoa from seminal plasma invariably require a centrifugation step. It has been suggested that the use of a cushioning medium formulated with a nonionic iodinated compound (iodixanol) during centrifugation ameliorates some of the damaging forces associated with the procedure, such that higher g-forces and longer durations of centrifugation may be used to maximize the yield of spermatozoa without deleterious effects on in vitro parameters [19,20] or fertility [21]. However, there is no compelling evidence to suggest that the use of a cushion during centrifugation results in improved sperm parameters using standard centrifugation protocols. To completely avoid the deleterious effects of centrifugation, filters have been developed for the removal of seminal plasma and the concentration of spermatozoa. These filters have the advantage of not requiring a centrifuge and therefore being advantageous under field conditions, improving motility parameters [22], and causing less damage to the plasma membrane while reducing bacterial growth rates in chilled sperm suspensions when compared to centrifugation techniques [23].

There is a long-standing paradigm that it is the nonviable or poor quality spermatozoa that generate the most ROS [24] and that these cells will accelerate the demise of the remaining sperm population during semen storage. Aitken et al have recently described a potential mechanism for this phenomenon through the discovery of a ROS generating enzyme in stallion spermatozoa which can exploit the amino acid substrates found in animal protein-based semen extenders, and in particular, egg yolk–based cryodiluents [25]. This L-amino acid oxidase is released primarily from dead spermatozoa and should these cells be left in the insemination dose during semen processing and storage, a precipitous decline in motility and viability will occur [25]. For these reasons, methods for the removal of dead and dying spermatozoa continue to be investigated and refined.

One such method is density gradient centrifugation, a technique which isolates cells on the basis of density to permit the selection of spermatozoa with improved classical parameters of functionality, such as morphology, motility, viability, and DNA integrity [26,27], along with increased fertility [28]. Interestingly, denser (presumptive "high quality") stallion spermatozoa have reduced expression of several mRNAs [29], the significance of which remains elusive given that a number of these downregulated mRNAs are positively associated with fertility in other species [30]. Nonetheless, given that significant correlations between the in vitro parameters of density gradient–isolated spermatozoa and pregnancy rates have been reported [31], there is evidence to suggest that it is the more dense sperm with reduced mRNA expression that do indeed participate in fertilization.

Download English Version:

https://daneshyari.com/en/article/2394355

Download Persian Version:

https://daneshyari.com/article/2394355

Daneshyari.com