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Lecture title: Gender Selection

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Summary: Gender Selection

I. Differences between X and Y-bearing spermatozoa:

Sperm sex selection methods all pre-suppose the existence of fundamental differences between X-and Y-bearing spermatozoa which can be exploited to enrich one population or the other. These differences include the size, density, motility and surface properties of X- and Y-bearing spermatozoa.

It has been experimentally proven that there is a **difference in the DNA content** of X- and Y-bearing spermatozoa due to their differential chromosome constitution, a theoretical argument for a difference in the size of X- and Y- bearing spermatozoa was advanced by some authors, although for many years no repeatable experimental evidence was presented to substantiate the theory. The issue by using the polymerase chain reaction (PCR) technique; Primers from the putative human testisdetermining gene (SRY) on the Y chromosome were used to discriminate human male DNA and **blood samples** (from male and female blood) with 100% accuracy.

Statistical analyses showed that the length, perimeter and area of the sperm head, and the length of the necks and tail, were greater in X- than in Y-bearing spermatozoa.

One of the assumptions that have been used to justify enrichment of X- and Y-bearing spermatozoa using different preparation procedures is that Y-bearing spermatozoa swim faster than X-bearing spermatozoa and have a greater ability to penetrate viscous solutions and the interfaces between viscous solutions. This hypothesis was invoked to explain enrichment of spermatozoa with Y bodies (putative Y-bearing spermatozoa) after passage of spermatozoa through albumin gradients. While a difference in the motility of the two sperm populations is often assumed, there is in fact no direct experimental evidence to support this contention.

II. Sperm separation techniques:

Sex pre selection methods can be divided into two general groups:-

1. Those that separate spermatozoa according to the physical or kinetic features.

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2. Those that rely on nuclear characteristics specific to either X or Y chromosome bearing spermatozoa.

These, in turn, can be divided into *in vivo* methods designed to produce optimal conditions for fertilization by either X or Y spermatozoa or *in vitro* separation methods used for separation X or Y chromosome bearing spermatozoa.

Spermatozoa separation techniques are based on different principles like **migration**, **filtration** or **density gradient centrifugation**.

1- Migration techniques are governed by the forward progression of spermatozoa 2- while density gradient centrifugation and filtration techniques are based on grouping spermatozoa motility and their preservation at phase boundaries and attachment to filtration matrices.

The pre fertilization spermatozoa separation techniques that influence the sex ratio of offspring must adhere to certain criteria:

- 1. Assure reliable separation of X and Y chromosome bearing spermatozoa in sufficient quantities.
- 2. Spermatozoa must be viable after separation and must be able to fertilize the ovum.
- 3. Fast, simple and cost-effective.
- 4. Isolate as much motile spermatozoa as possible.
- 5. Eliminate toxic or bioactive reactive oxygen species (ROS).

Given that spermatozoa separation techniques require these criteria, a variety of spermatozoa separation techniques are mandatory in clinical practice to obtain the best possible acquiesce of functionally competent spermatozoa for insemination purposes.

The ideal sperm separation technique should be:

- 1- Quick, easy and cost-effective
- 2- Isolate as much motile spermatozoa as possible
- 3- Not cause sperm damage or non-physiological alterations of the separated sperm cells
- 4- Eliminate dead spermatozoa and other cells, including leukocytes and bacteria
- 5- Eliminate toxic or bioactive substances like decapacitation factors or reactive oxygen species (ROS)
- 6- Allow processing of larger volumes of ejaculates.

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Since none of the methods available meets all these requirements, a variety of sperm separation techniques is mandatory in clinical practice to obtain an optimal yield of functionally competent spermatozoa for insemination purposes. Depending on the ejaculate quality, these methods have different efficiency and areas of use.

The most important spermatozoa separation techniques that are currently being used are the following:-

- **1-** Albumin separation technique.
- **2-** Density gradient centrifugation.
- **3-** Swim-up procedure.
- **4-** Flow cytometry.
- **5-** Migration-sedimentation.
- **6-** Glass wool filtration. **7-** Free Flow Electrophoresis **8-** Sephadex columns.
- 9- Tran's membrane migration.