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	Effective Date:	06-30-2022
	Document Type:	Standard Operating Procedure
	Page Number:	1 of 4
	Department/Section:	Hematology
	Document Title:	SPERM WASHING

PURPOSE:

To establish guidelines in collecting sample for Sperm Washing for Laboratory diagnostic procedure and analysis. It is used in artificial insemination using the intrauterine insemination (IUI) technique and in vitro fertilization (IVF). It may also be used to decrease the risk of HIV transmission by an HIV-positive male, in which case the washed sperm is injected into a female using an artificial insemination technique.

SCOPE:


Applies to all Hematology Section Staff of Laboratory Department of Dr. Pablo O. Torre Memorial Hospital (DPOTMH)

PERSON RESPONSIBLE:

Medical Technologists (Medical Laboratory Scientists), Pathologists, Medical Doctors, Nurses, Medical Trainees, Laboratory Clerks

GENERAL GUIDELINES:

1. Specimen maybe collected at the laboratory collection room or maybe immediately brought in by the client right after collection outside the laboratory. If the client is on site, he shall notify the laboratory regarding the time for the sample to arrive.
2. The sample shall arrive in the laboratory within 1 hour of collection for appropriate analysis. It is critical for the specimen to be examined while it is fresh. Sperm motility is the most affected by this time limitation, but the other values are not affected. It shall be indicated in the laboratory slip the time sample is collected and received.
3. Semen specimens shall be collected in a clean glass or non-toxic plastic, dry, wide mouth container. The entire specimen is needed for analysis. (Analysis of ejaculate changes from initial secretions and the later secretions. Accuracy cannot be ensured if the entire sample is collected).
4. Masturbation shall be the preferred method of collection. However, this method may not acceptable to some clients. Specifically manufactured condoms, which are free of spermicides may be available. Coitus interruptus is least acceptable because

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	Effective Date:	06-30-2022
	Document Type:	Standard Operating Procedure
	Page Number:	2 of 4
	Department/Section:	Hematology
	Document Title:	SPERM WASHING


B.S. Aquino Drive,
Bacolod City,
Negros Occidental,
6100

some of the initial ejaculate may be lost and the sample may be contaminated by vaginal secretions.

5. If the sample is collected off site, it shall be transported at near body temperature to ensure accurate result (Note: keeping the collected sample collected inside the shirt and close to the body will help maintain the temperature). Instruct the client to avoid excessive heat or cold when transporting the specimen.
6. An accurate history of prescription and recreational drugs, habits (such as regular hot tubs), injuries and infections is important.
7. Adhere to standard precautions. Provide adequate privacy for the collection of the specimen.
8. Turn-around time for releasing Sperm Washing result is 4 hours.

PROCEDURE:


1. The Medical Technologist examines the gross appearance of the sample noting the color, pH reaction, volume and the liquefaction on viscosity freshly ejaculated semen is a viscid, opaque, white coagulum. Normally the volume ranges from 1.5 to 5 mL. Infertile males often have an increased volume with a diminished sperm count.
2. Stand semen sample for 30 minutes to 1 hour at room temperature to liquefy and record Pre motility and Pre sperm count.
3. To evaluate the sperm motility, the Medical Technologist places a drop of seminal fluid. The Medical Technologist examines the specimen under high power objective to determine the percentage of motile spermatozoa. Normally more than 70% of spermatozoa show active motility within one hour of collection. In addition, the numbers of white and red blood cells are also noted.
4. The Medical Technologist puts into the pipette the well-mixed liquefied seminal fluid to 5 marks and dilute to 11 mark of the WBC pipette.
5. The Medical Technologist loads the specimen into the pipette shaker to vigorously mix.
6. The Medical Technologist charges the specimen into a Neubauer hemacytometer chamber, allows it to settle for 2 minutes. Counts four large corner squares of the

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	Effective Date:	06-30-2022
	Document Type:	Standard Operating Procedure
	Page Number:	3 of 4
	Department/Section:	Hematology
	Document Title:	SPERM WASHING

- counting chamber and multiplies the figure obtained by 50,000 to obtain 60-150 million count per mL normal range from per mL are associated with infertility.
- The Medical Technologist notes down the sperm morphology by percentage of normal and abnormal forms from the stained pull-apart smear.
 - Into a sterile conical tube, add 2.5 mL and 5 mL of flushing medium, 1:2 parts.
 - Mix well. Centrifuge at 1600 for 15 minutes. Remove supernatant and leave 1 mL pellet. Mix pellet.
 - Gently overlay 2 mL of flushing medium into the sperm pellet. Incubate at 37C for 45-60 minutes (BB incubator).
 - After incubation, cautiously draw 1 mL of the top layer without disturbing the pellet and transfer into a sterile 5 mL test tube.
 - Dilute this with 2 mL flushing medium. Centrifuge at 1600 rpm for 5 minutes and discard supernatant. Resuspend the sperm pellet in 1 mL flushing medium.
 - Assess the recovered sample and record Post sperm count and Post motility.
 - The Medical Technologist notes all macroscopic and microscopic findings and encodes results in the system.
 - The Medical Technologist/Laboratory Clerk prints out results for outpatients at the reception area upon presentation of the Official Receipt (OR) and results for admitted patients can be viewed and printed out by nurses to its respective stations.
 - Names and results of patient shall be recorded in the Hematology/Miscellaneous logbook.


REFERENCE:

- <https://www.webmd.com/infertility-and-reproduction/guide/semen-analysis>


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	Page Number:	4 of 4
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	Effective Date:	06-30-2022
	Document Type:	Work Instruction
	Page Number:	1 of 2
	Department/Section:	Hematology
	Document Title:	SPERM WASHING

KEY TASKS	PERSON RESPONSIBLE
1. Encodes the request of the patient to BIZBOX system.	Laboratory Clerks and Nurses
2. Instructs patient for sample preparation.	Medical Technologist/Nurse
3. Provides adequate privacy for the collection of the specimen.	
4. Ensures sample is acceptable for testing.	
5. Encodes the request in the system.	
6. Processes and analyzes samples.	
7. Releases and validates result thru BIZBOX System.	
8. Records result to Hematology logbooks.	
9. Endorses the patient's results to the patient or doctor.	Laboratory Clerks and Nurses

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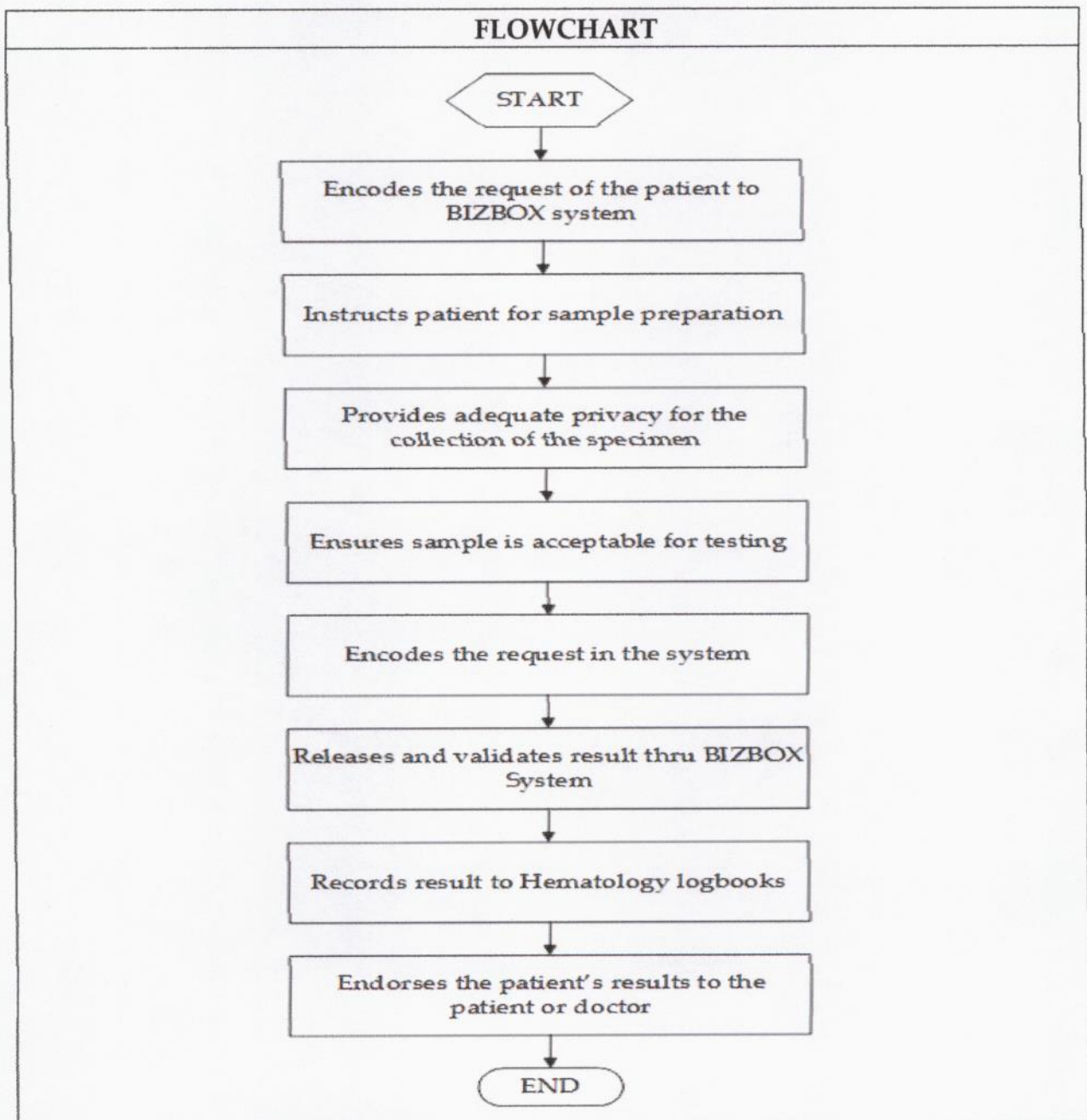



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Document Code:	DPOTMH-E-57-P01-FC24
Effective Date:	06-30-2022
Document Type:	Flowchart
Page Number:	1 of 2
Department/Section:	Hematology
Document Title:	SPERM WASHING

FLOWCHART



 <p>DR. PABLO O. TORRE MEMORIAL HOSPITAL</p>	Document Code:	DPOTMH-E-57-P01-FC24
	Effective Date:	06-30-2022
	Document Type:	Flowchart
	Page Number:	2 of 2
	Department/Section:	Hematology
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