PROTOKOL - EMBRYOLOGY- PRACTICALS 2 NAME:

Methods of andrological analysis

24.2.2022

THEORY

- sperm count test: basic examination of a semen sample evaluating male fertility
- basic values of native semen in men:

total sperm count (mil./ejaculate)	39 mil.
liquefaction	do 60 minut
pH of liquefied sample	7,2-7,8
sperm concentration	15 mil./ml
vitality (eosin staining)	58 %
total motility	40 % (progressive + non progressive)
progressive motility (linear)	32 %
normal morphology (head, neck and tail morphology)	4 %
presence of white blood cells, prokaryotic cells	

- Note: reference values in 2009 for men: 20 mil./ml, motility 50 % (25 % progressive motility), morphology 30 %,
  reference values in 1960: 80 mil./ml

- basic values of native semen in pigs (*Sus scrofa forma domestica*):

pH	7,2
sample volume	> 100 ml
sperm concentration	> 150 mil./ml
total motility	> 70 %
abnormal sperm morphology inc. cytoplasmatic droplets	< 25 %

- basic findings and abnormalities:

normozoospermia	see reference values
oligozoospermia	low sperm count
asthenozoospermia	reduced sperm motility
teratozoospermia	pathological shapes
cryptozoospermia	less than 1 mil./ml
azoospermia	semen contains no sperm cells
nekrozoospermia	dead sperm cells
OAT syndrome	oligo-astheno-teratospermia

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Picture 1: Abnormalities of sperm cells:

	Morphologically normal	34
Head Anomalies	Tapered	0
	Thin	3
	Microcephalous	3
	Macrocephalous	2
	Multiple	0
	Abnormal post-acrosomal region	26
	Abnormal or absent acrosome	53
Midpiece Anomalies	Cytoplasmic droplet	2
	Thin	0
	Bent	3
Tail Anomalies	Absent	2
	Short	0
	Irregular	0
	Coiled	9
	Multiple	0
	Total number of isolated and associated anomalies = **T**	103
	✦ **Multiple Anomalies Index (MAI)** = **T** / number of abnormal sperm (66 for 100 spermatozoa assessed)	1.56

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PRACTICAL PART

- **sample evaluation using Bürker chamber (BCh):**

a) sample dilution: mix 50 µl of the sample with **.....** µl of buffered physiological saline solution in a test tube. Mix the sample well and apply under the cover glass of the Bch. Wait 3 minutes. Then evaluate the sample under the light microscope using the magnification from 200 x to 400 x. **Evaluate the sample in 10 squares of Bch, unless otherwise indicated**. Count the cells inside the square and on 2 sides (see graphic materials).

b) concentration assessment (c): count motile and immotile cells separately, then calculate *c*:
c = (total number of counted cells x dilution) / (number of squares x square area x heigh of Bch)

c) motility assessment:

I) total motility in %:

total motility in % = c of motile sperm cells / total c

II) % of cell with progressive motility: evaluate min. 100 cells (optimally 200 cells), calculate % of cells with progressive movement

d) morphology evaluation: evaluate min. 100 cells (optimally 200 cells), calculate % of cells with wi normal morpsholgy, calculate abnormalities of head, neck and tail separately

e) record the presence of prokaryotic cells white blood cells and ather abnormalities

CONCLUSION

Date	Concentration	Motility	Normal morphology

REFERENCES

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