NAMIBIA UNIVERSITY
OF SCIENCE AND TECHNOLOGY

Faculty of Health and Applied Sciences

Department of Health Sciences

QUALIFICATION: **BACHELOR OF MEDICAL LABORATORY SCIENCES**	
QUALIFICATION CODE: **08BMLS**	LEVEL: **6**
COURSE: **MOLECULAR DIAGNOSTICS**	COURSE CODE: **MOD621S**
DATE: **NOVEMBER 2019**	SESSION:
DURATION: **3 HOURS**	MARKS: **100**

FIRST OPPORTUNITY EXAMINATION QUESTION PAPER	
EXAMINER(S)	Ms V Tjijenda
MODERATOR:	Dr A Shiningavamwe

INSTRUCTIONS

1. Answer all questions.
2. Please write neatly and legibly.
3. Do not use the left side margin of the exam paper. This must be allowed for the examiner.
4. No books, notes and other additional aids are allowed.
5. Mark all answers clearly with their respective question numbers.

Permissable material

Non programmable calculator is allowed.

THIS QUESTION PAPER CONSISTS OF 3 PAGES (Excluding this front page)

SECTION A (27 MARKS)

QUESTION 1 **[15]**

Write short notes on the following.

1.1 Sanger sequencing method (3)

1.2 Taq polymerase (3)

1.3 Diethyl pyrocarbonate (DEPC) (3)

1.4 Uni-directional work flow (3)

1.5 Central dogma of Biology (3)

QUESTION 2 **[12]**

Identify ONE assay that can be used for each of the following:

2.1 Compare gene expression in acute myeloid leukemia and chronic myeloid Leukemia. (1)

2.2 Amplify DNA sequence to make multiple copies. (1)

2.3 Locate the chromosome number responsible for down syndrome. (1)

2.4 Convert viral RNA to cDNA for further analysis. (1)

2.5 Quantification of viral load. (1)

2.6 Proteomic analysis. (1)

2.7 Separation of DNA and RNA based on charge and size. (1)

2.8 Genomic strain typing of an *E. coli* outbreak. (1)

2.9 Identification of novel mutations. (1)

2.10 Forensic investigations. (1)

2.11 Modified form of polymerase chain reaction (PCR) which avoids a non-specific amplification of DNA by inactivating the DNA polymerase at lower temperatures. (1)

2.12 NGS technology that sequence DNA via three basic processes: amplify, sequencing and analyses using a bridging method. (1)

SECTION B (43 MARKS)

QUESTION 3 [10]

3.1 Define restriction enzyme. (1)

3.2 Design a 10 nucleotides long palindrome sequence. (3)

3.2.1 Digest the palindrome sequence obtained in 3.2 such that it yields a blunt end. (2)

3.2.2 Digest the palindrome sequence obtained in 3.2 such that it yields a 3' sticky end. (2)

3.3 Provide the formula for calculating melting temperature. (2)

QUESTION 4 [7]

Using your knowledge of nucleic acid extraction and purification using the phenol chemical method, answer the following questions.

4.1 Identify four components of the lysis buffer. (4)

4.2 Explain the importance of the chloroform/isomamylalcohol (24:1) step. (1)

4.3 Why is sodium acetate added. (1)

4.4 Explain the role of ice-cold isopropanol. (1)

QUESTION 5 [26]

Neisseria gonorrhoeae is a sexually transmitted infection with resistance to previously and currently recommended antimicrobials. Both culture and Southern blotting technique are used for diagnosis. The *ctaA* gene encodes an outer membrane protein that's a target for antibiotics and is used as target for PCR. The presence of the *ctaA* gene confers antibiotic resistance. Three cases, A, B, and C of suspected *Neisseria gonorrhoeae* gave the following results during diagnosis:

CASES	**A**	**B**	**C**
DST Culture	**S**	**R**	**R**
Southern Blotting (gene *ctaA*)	**-**	**-**	**+**

Table 1: DST culture and Southern Blotting drug resistance results for *N. gonorrhoeae.*

Provide the likely explanations for either the discrepancy or congruency between the DST culture and northern blotting results.

5.1.1 For patient A (2)

5.1.2 For patient B (2)

5.1.3 For patient C (2)

5.1.4 From your observation, what does it suggest on the sole use of *ctaA* detection in *N. gonorrhoeae* resistance diagnosis. (2)

5.1.5 Is this molecular method quantitative or qualitative. Justify. (2)

5.2 Explain the steps in the Southern Blotting method. (10)

5.3 Compare traditional PCR to real time PCR. (6)

SECTION C (30 MARKS)

QUESTION 6

6.1 Discuss the principle of Western Blotting. (10)

6.2 You are employed at NSVP Scientific molecular department. You are requested to design primers for a postgraduate master's research. Explain important considerations when designing the primers. (10)

6.3 Generate the gel electrophoresis profile of the following sequence using the Maxam Gilbert chemical method. (10)

5' ATTGACTTAGCC 3'

END OF EXAMINATION