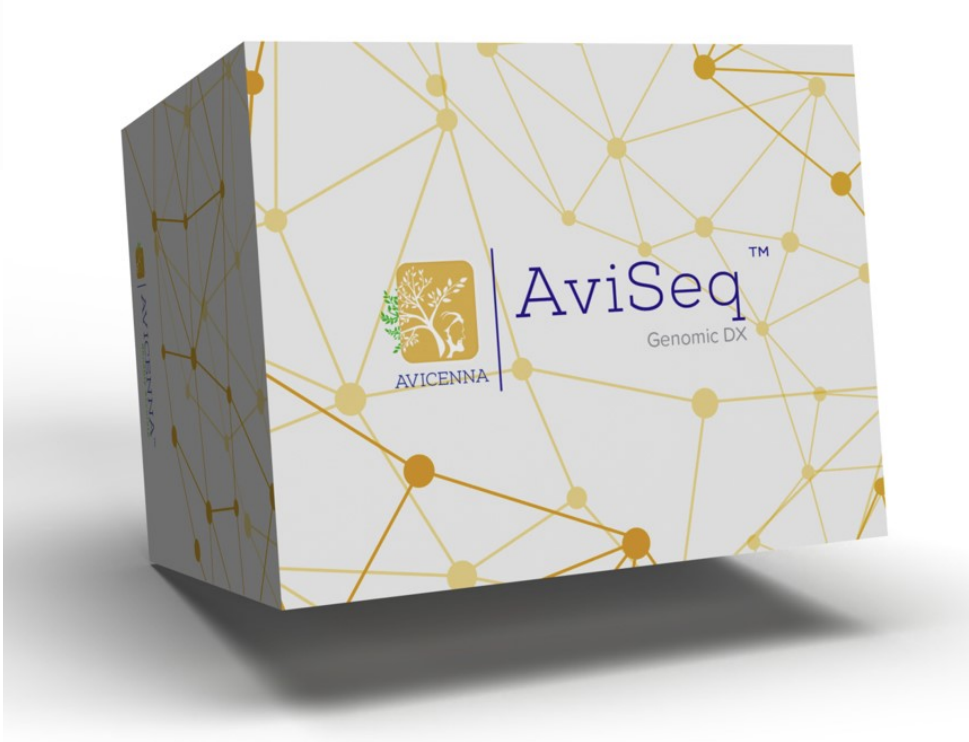
AVICENNA

AviSeq™

Genomic DX

Barcode Set 1-16

REF: AVG502 96 TESTS



USER GUIDE

AVG504_IFU_Rev00_MAY22

CE
IVD

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1. PRODUCT APPLICATIONS

Aviseq Barcode Set 1-16 provided in this kit must be used in combination with the AviSeq range of kits to prepare libraries for next-generation sequencing on the Ion Torrent range of sequencing instruments.

2. KITS CONTENTS

Aviseq Barcode Set 1-16 kit contains ready-to-use reagents for the insertion of the barcodes and adapters to DNA libraries rendering them suitable for analysis on Ion Torrent instruments. The PCR product from Step 1 of the Target Enrichment protocol is the starting material (DNA) for this PCR, in which Barcodes and Adapters are added to the PCR products to produce a library suitable for NGS on the Ion Torrent instruments only.

Aviseq BARCODE SET 1-16 (cod. AVG502)			
TUBE	**VOLUME**	**TUBE**	**VOLUME**
Barcode 1	24 µl	Barcode 9	24 µl
Barcode 2	24 µl	Barcode 10	24 µl
Barcode 3	24 µl	Barcode 11	24 µl
Barcode 4	24 µl	Barcode 12	24 µl
Barcode 5	24 µl	Barcode 13	24 µl
Barcode 6	24 µl	Barcode 14	24 µl
Barcode 7	24 µl	Barcode 15	24 µl
Barcode 8	24 µl	Barcode 16	24 µl

Table 1: Content of the Aviseq Barcode Set 1 16 kit.

3. STORAGE AND STABILITY

All reagents provided in this kits are ready to use and must be stored at -20°C.
A high standard of performance capacity and quality will be maintained until the expiry data marked on each reagent tube and on the external jar/container if the kit remains intact, is properly stored and undergoes a maximum of 3 freeze-thaw cycles.
When the reagents are thawed they must be kept on ice throughout the process and returned to -20°C storage after use immediately.

4. REQUIRED MATERIAL NOT INCLUDED

4.1 Generic Material

- Computer with constantly updated and guaranteed secure internet connection
- Micropipettes calibrated and periodically verified 0.2-2 µl, 2-20µl, 20-200µl or 100-1000µl and filter tips
- Vortex
- Disposable Gloves without powder and other appropriate PPE.
- Thermal cycler calibrated and periodically verified
- Tubes and Caps or 96-wells plate, as needed, DNase and RNase free
- Nuclease-free water
- 1,5 ml tube magnetic separator or 96-wells plate compatible magnetic separator
- Fresh 70% ethanol and 5% hypochlorite
- Ion Torrent sequencer calibrated and periodically verified

4.2 Specific Material

The material listed below has been validated for use in this procedure and with the reagents provided in this kit:

- Qubit™ 2.0 Fluorometer (Invitrogen Cod. Q32866) or Qubit™ 3.0 Fluorometer (Invitrogen Cod. Q33216) or Qubit™ 4.0 Fluorometer (Invitrogen Cod. Q33226) calibrated and periodically verified
- Qubit™ assay tubes (Invitrogen Cod. Q32856)
- Qubit™ dsDNA HS Assay Kit (Invitrogen, cod. Q32851)
- Magnetic beads for the purification of genomic libraries, AMPure XP (Beckman Coulter, cod. A63880)

QUALITATIVE ANALYSIS OF DNA (Optional)

- Agilent 2100 Bioanalyzer system with DNA reagent kit calibrated and periodically verified

5. IMPORTANT NOTES AND SAFETY INFORMATION

The Users must comply with the recommendations listed below and failure to do so will result in suboptimal outcomes or failed runs. If the kit or any component thereof is forwarded to a third party, it is the your responsibility to provide the end user with these recommendations. Note: the manufacturer is committed to the highest level of quality assurance at all steps in the procedure and will provide full support to all customers.

- This kit is intended for use by professionals trained in molecular biology. Use by untrained individuals may result in suboptimal outcomes.
- Do not use if package damaged
- Biological samples and all reagents should be handled in properly equipped rooms, clean and clear of potential contaminants. We highly recommend cleaning working areas frequently using a solution containing sodium hypochlorite 1-5% followed by sterile water and 70% Ethanol.
- Always use appropriate personal protective equipment (PPE) such as laboratory coat, gloves and safety goggles during all steps described in the protocol.
- Ensure that you are aware of all safety instructions associated with all equipment and related electrical supply, chemicals and other resources utilized in the performance of the procedures outlined in this IFU.
- Ensure that the quality control procedures in place at each phase of the protocol are adequate to detect any errors, particularly after each usage of the kit. This should include a quality evaluation of the results generated. Contact your local supplier immediately on detecting any errors and/or anomalies in the performance of the kit.
- It is essential in a diagnostic and/or clinical setting that users of this kit put strong control and quality assurance procedures in place to minimize or eliminate diagnostic errors following a critical evaluation of possible risk factors in the protocol. Any errors detected must be reported to the relevant medical staff immediately.
- To avoid contamination of reagents we recommend using DNase/RNase free tubes, filter tips and to pay particular attention to keep all instruments clean and free of contaminants.
- We highly recommend the lab is designed in a unidirectional workflow from the initial phase of DNA isolation following the PCR preparation phase, amplification and post-amplification phases in order to keep working areas separated for the different phases of the procedure. Each area must have dedicated laboratory equipment (e.g., vortex, pipettes, etc.,), consumables (e.g. tubes, pipette tips, etc.,) and PPE (e.g., gloves, etc.,).
- Waste reagents and biological samples must be discarded according to local regulations and legal procedures.

6. PROTOCOL

For detailed instructions on the use of the barcodes provided in this kit, please refer to the instructions for use of the the AviSeq kit you are utilizing.

7. TROUBLESHOOTING

PROBLEM	POSSIBILE CAUSE	SUGGESTION
Absence of bands on agarose gel after electrophoresis	Incorrect thermocycling profile	Verify the PCR thermal profile and calibration then repeat the PCR reaction
	Mistakes in master mix preparation	Verify PCR mix components and repeat the PCR reaction
	Degraded reagents	Verify expiry date and storing conditions of the products
	Presence of inhibitors	Verify concentration and quality of DNA extracted using a spectrophotometer. If necessary, repeat DNA extraction.
	Low DNA concentration	Verify concentration and quality of DNA extracted using a bioanalyser. If necessary, repeat DNA extraction.
Absence of bands on agarose gel after electrophoresis	Wrong PCR machine settings	Verify the PCR thermal profile and calibration then repeat the PCR reaction
Presence of fragments with low molecular weight	Mistakes in master mix preparation	Verify PCR mix components and repeat the PCR reaction

8. SYMBOLS

CE	*According to 98/79/CE Directive*	REF	*Catalogue number*
IVD	*In Vitro Diagnostic Medical Device*	LOT	*Batch code*
	*Expiration date*		*Temperature limitation*
	*Consult instruction for use (IFU)*		*Sufficient for n. tests*
	*Manufacturer*		*Do not use if package damaged*