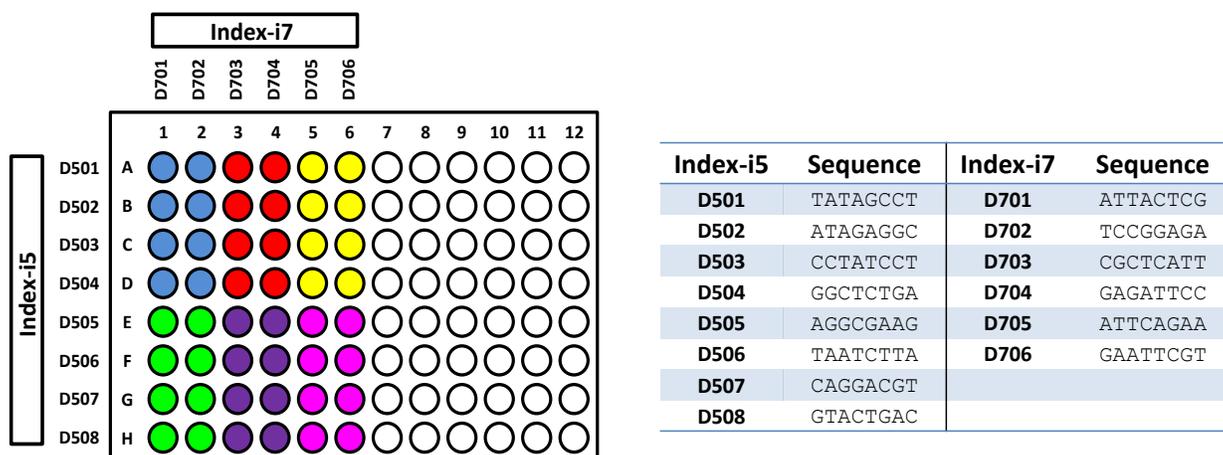


## Dual Index Plate Map, Sequences, and Instructions for Use

PicoPLEX DNA-seq Dual Index Plate contains 48 different dual index combinations in a single use format in a 96-well plate. Since the **benefit of the use of dual barcodes is to ensure sample identity**, it is critical to handle these indexes **with care** to ensure that there is no barcode cross contaminations and related problems. Please follow the instructions below:

1. Thaw plate for ten minutes bench-top prior to use.
2. After thawing, spin plate in table top centrifuge to ensure contents are at bottom of well.
3. Thoroughly wipe foil seal with 70% ethanol. Allow to dry.
4. Pierce seal above target index with 200  $\mu$ L or 1.0 mL clean pipet tip.
5. Discard tip used for piercing.
6. Use clean appropriate pipet to collect 5  $\mu$ L of a specific index and add to the reaction at amplification step.
7. Cover any pierced/used index wells with scientific tape to mark the one-time usage index as used. (Tape such as VWR General Scientific Tape 0.5" Cat# 89097-920)
8. After index plate is used, thoroughly wipe seal with 70% ethanol. Dry completely.
9. Replace plastic lid, return the plate to its sleeve and store at  $-20^{\circ}\text{C}$ .
10. Avoid repeated freezing and thawing of the plate. **Do not exceed** 4 freeze/thaw cycles.

**Note:** The kit contains Illumina dual barcode oligonucleotides, and these indexes can't be substituted with index oligonucleotides from any other sources.



### Dual index plate map with well locations:

Single use index plate representing well locations (left) of 48 dual indexes, with appropriate i7 (columns) and i5 (rows) index combinations; the sequences of respective indexes are shown in the table (right). Wells A7-H12 are blank wells and do not contain any index oligonucleotides. **(Figure-2 is repeated here for convenience) Any alternate combinations of indexes should be validated with Illumina Experiment Manager to ensure sequencing diversity.**

**Handling for low throughput applications: If the entire Dual Index Plate is not used, it is critical to follow the instructions below to avoid cross contamination.**

1. After removing indexes of choice, cover any pierced or used index wells with scientific tape such as VWR General Scientific Tape 0.5" Cat# 89097-920.
2. Thoroughly wipe the seal with 70% ethanol and allow it to dry completely.
3. Replace the plastic lid, return the Dual Index Plate to its sleeve and store at  $-20^{\circ}\text{C}$ .