Specifications

Power	Supply voltage	24 VDC, 6 A
	Adapter input	100-240 V~, 50/60 Hz, 250 VA
Dimensions	356 mm (W) ×257.5 mm (H)×315 mm (D) (14 inches ×10 inches×12 inches)	
Net weight	Approximately 15 kg (33 lb)	
Touch screen	Type	LCD
	Size	8 inches
	Resolution	1024×600 pixels
Maximum sound pressure level	65 dBA	
Lab bench load-bearing capacity	30 kg/m^2	
Operating environment requirements	Temperature	15°C to 30°C (59°F to 86°F)
	Relative humidity	20% to 60%. non-condensing
	Atmospheric pressure	70 kPa to 106 kPa
	Maximum altitude	1800 m (5906 ft)
Storage/transportation environment requirements	Temperature	-20 °C to 50°C (-4°F to 122°F)
	Relative humidity	15% to 85%. non-condensing
	Atmospheric pressure	70 kPa to 106 kPa
	Maximum altitude	3000 m (9843 ft)
Accompanying items	Refer to the packing list	

Fully enclosed Library Prep
One Cartridge for All Steps

DNBelab-D4

Digital Sample Preparation System

MGI Tech Co., Ltd.
Building 11, Beishan Industrial Zone, Yantian District, Shenzhen, CHINA
MGI-service@mgi-tech.com en.mgi-tech.com +86-4000-688-114

Version number: August 2024 version I MGPC70003001

DNBelab-D4
Digital Sample Preparation System

DNBelab-D4 utilizes a fully enclosed digital microfluidic technology and integrates multiple function modules in one instrument. It can perform all library preparation steps, including enzymatic fragmentation, end repair, adapter ligation, PCR, magnetic bead purification, fragment size selection, nucleic acid quantification, DNB making, etc. on one cartridge in a fully enclosed and automated manner. DNBelab-D4 enables loading nucleic acids (DNA and/or RNA) into cartridge and taking out the final product (double-stranded DNA library or DNB) while providing the concentration of the product and the recommended amount of input for sequencing.

DNBelab-D4 Instrument

- The user interface is on the touchscreen and there is no need for an extra computer
- The instrument integrates a cryopreservation module, a temperature control module, a quantification module, a magnet module and a droplet manipulation module, etc.
- The instrument with a 0.1 m^2 footprint and <15 kg weight is portable and easy to deploy

Cryopreservation Module

Enable low-temperature storage function for reagent reservoir zone on sample preparation cartridge

Temperature Control Module

Achieve precise temperature control with rapid thermal cycling to conduct biochemical reactions such as PCR

Quantification Module

Achieve quantification of nucleic acid

Droplet Manipulation Module

Achieve droplet dispensing with determined volume, droplet moving, merging, mixing, etc.

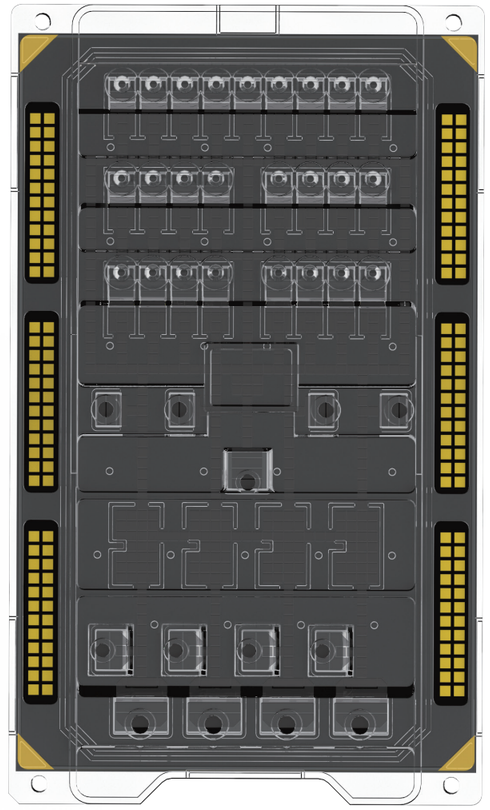
Magnet Module

Achieve magnetic beads-based nucleic acid purification and fragment size selection

* DNB: DNA Nanosphere, DNA nanoballs

DNBelab-D4 Sample Preparation Cartridge

- Sealing fluid, samples and all reagents are loaded into the cartridge. One cartridge can achieve all library prep steps in a fully enclosed manner
- All the waste generated during library preparation is also retained and sealed in the sample preparation cartridge. Extra steps of waste treatment are not required.
- The cartridge supports 1-4 samples per run.

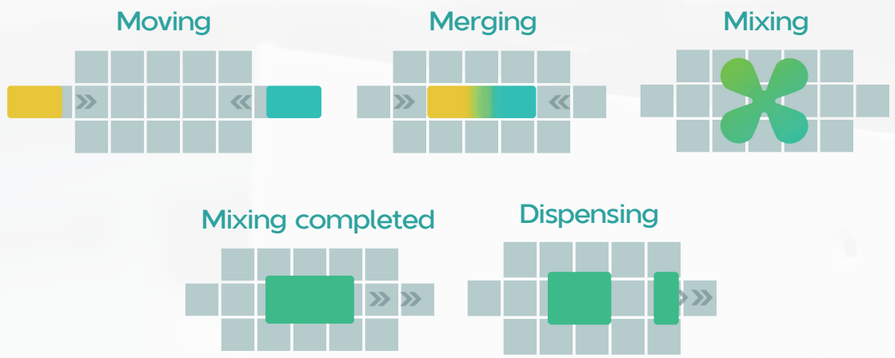


The installation of DNBelab-D4

After unpacking the DNBelab-D4 and placing it on the desktop, leveling the D4 instrument with the leveling device included in the package is required, which usually takes less than 5 minutes. The setup is completed, and it can be used immediately. If the D4 instrument is moved, it needs to be leveled again and then used.

DNBelab-D4
Performance

DNBelab-D4 utilizes digital microfluidic technology based on electrowetting to control the microliter-level droplets to perform moving, merging, mixing, dispensing and other operations on the sample preparation cartridge. These droplet operations cover all the liquid manipulations required in the library preparation, including reagent dispensing with determined volume, mixing of different kinds of reagent, liquid transfer, etc. DNBelab-D4 can be used for whole genome sequencing, metagenomics sequencing, targeted sequencing, library conversion, etc.

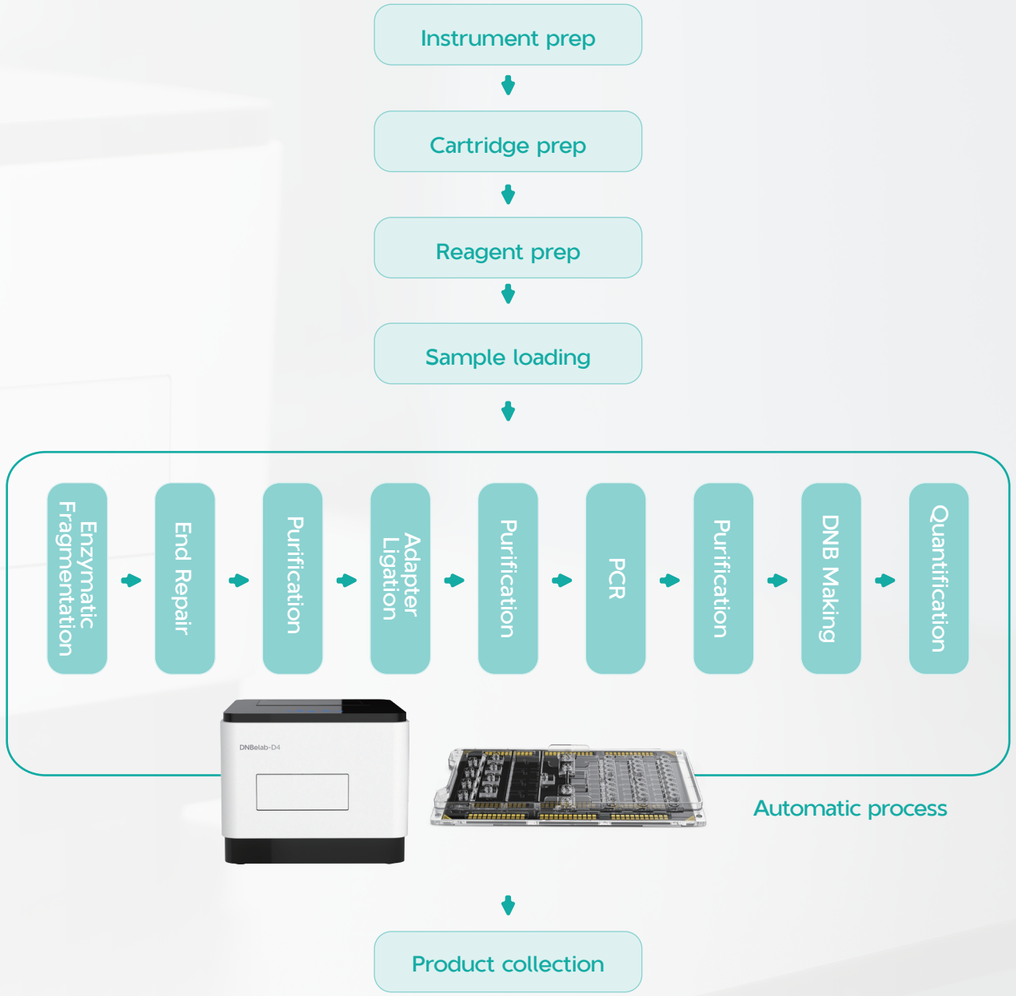


Technology	Fully enclosed digital microfluidic technology
Throughput	1-4 samples per run
Hands-on time	< 15 mins (including reagent preparation, sample loading, etc.)
Certification	NMPA, CE-IVD, CE-RUO, NRTL and EAC
Input	Extracted DNA or/and RNA
Sequencer	DNBSEQ-E25, DNBSEQ-G99, DNBSEQ-G50, DNBSEQ-G400 and so on
Compatibility for third-party kit	Support customized development of third-party kits

* Customization: MGI supports the customized development of third-party kits.

Compared with manual library preparation that requires several instruments with different functions, DNBelab-D4 integrates multiple function modules in one instrument, which saves time for manual transfer between different instruments. Therefore, it can achieve faster library preparation. It takes 2.5 h to achieve DNA in and DNB out with quantification results by using DNBelab-D4RS fast PCR-FREE FS Library Prep Kit.

Moreover, DNBelab-D4 supports the customized development of the third-party kit. The library preparation steps that can be done on DNBelab-D4 include reagent pre-mixing, reverse transcription, enzymatic fragmentation, end repair, A-tailing, adapter ligation, PCR (targeted amplification), magnetic bead purification, fragment size selection, nucleic acid quantification, DNB making, etc.

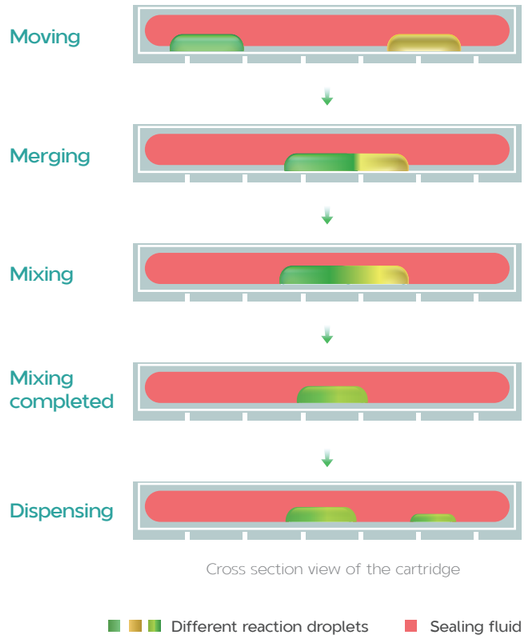


Taking library preparation with enzymatic fragmentation (including PCR) for WGS as an example. The actual steps depend on the specific library preparation method.

DNBelab-D4

Fully enclosed library prep
One cartridge for all steps

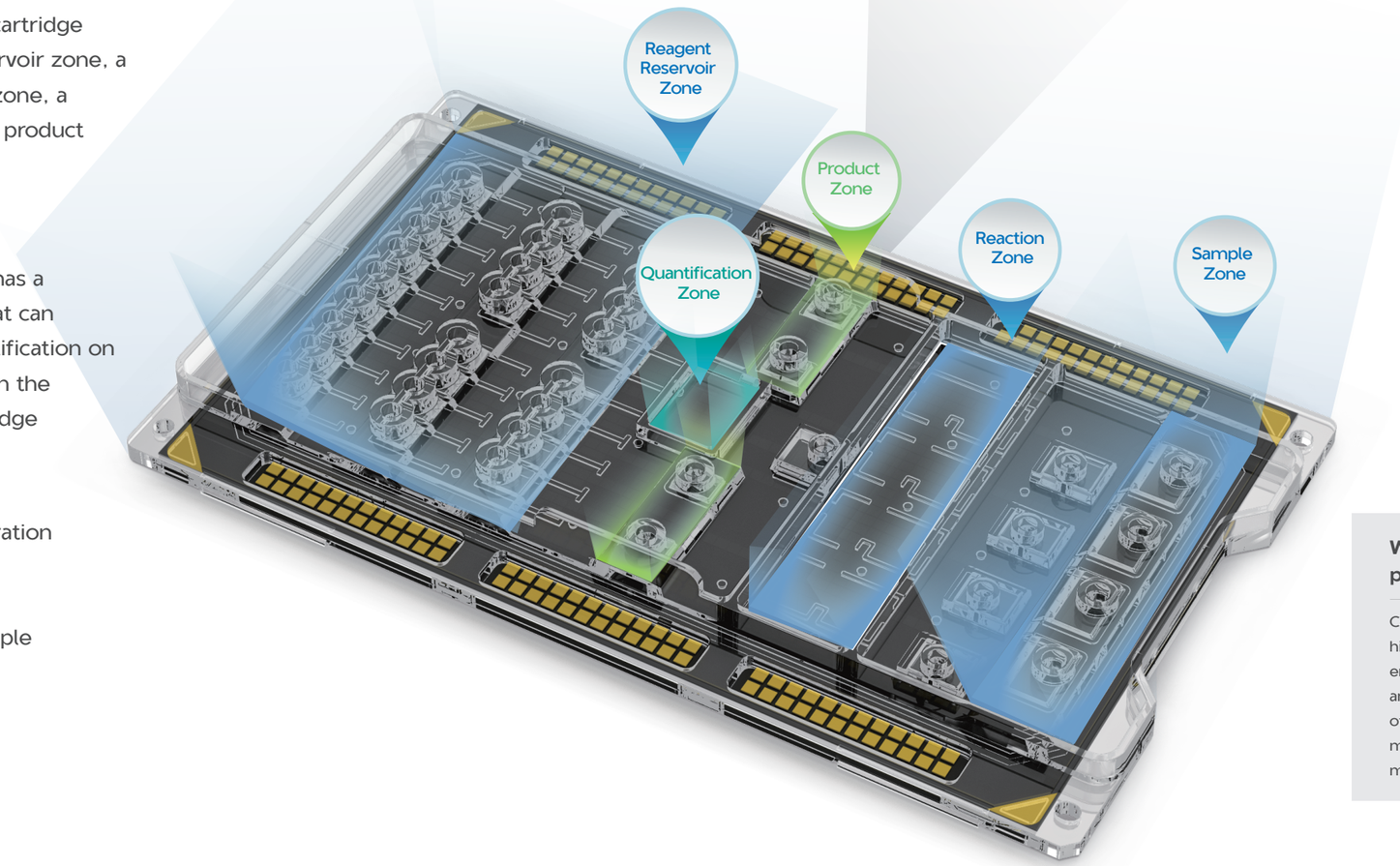
DNBelab-D4 utilizes a unique fully enclosed system. In the process of library preparation, every droplet is always wrapped by the sealing fluid while all the waste is also wrapped by the sealing fluid and retained in the sample preparation cartridge. These droplets are completely isolated from the ambient air, so no aerosol is produced. Each reaction zone is independent of the other, thus completely avoiding aerosol contamination and sample cross-contamination. The prevention of cross-contamination has been validated for mNGS and tNGS for pathogen identification, small oncology panel, and other sequencing applications, and the results show that D4 can achieve zero-contamination library preparation.



- The sample preparation cartridge integrates a reagent reservoir zone, a sample zone, a reaction zone, a quantification zone and a product zone

- DNBelab-D4 instrument has a quantification module that can achieve automated quantification on the quantification zone on the sample preparation cartridge

- All steps of library preparation for high-throughput sequencing can be done automatically on the sample preparation cartridge



What are the advantages of fully enclosed library preparation?

Compared with PCR test, mNGS has higher coverage for pathogen and higher requirement for prevention of aerosol contamination. While ensuring the quality of experiment, it is preferred to use fully enclosed and automated instrument with confirmed performance to replace part of manual operation, which is beneficial to prevent contamination of microorganism or nucleic acid from operator and laboratory environment during the experiment.

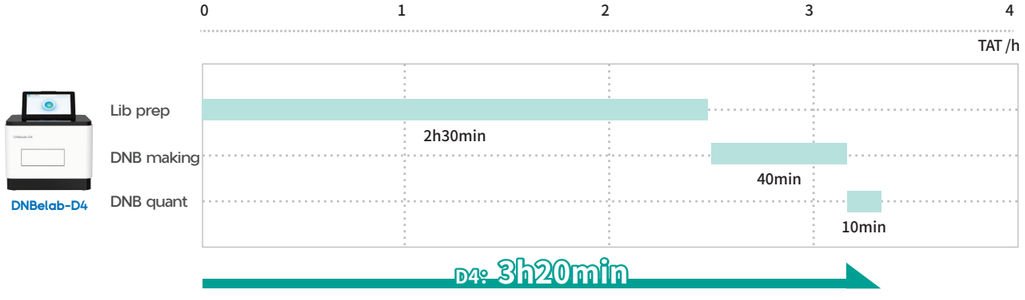
Case 1: Small Whole Genome Sequencing

Experiment Scheme

- **Sample:** microbial DNA
- **D4:** DNA in, DNB out with DNB quant result
- **Library prep Kit：** DNBelab-D4RS FS DNA Library Prep Set V2.0
- **D4 TAT:** 3h20min (Including DNB making and DNB quantification)
- **Sequencing:** DNBSEQ-E25 PE150+10+10
  DNBSEQ-G99 PE150+10+10
- **Bioinformatics:** MGAP

Conclusions

- DNBelab-D4 can obtain DNB with sufficient concentration (≥10ng/μL)
- Both DNBSEQ-E25 and DNBSEQ-G99 can generate sufficient data amount with high quality for analysis
- All bacteria in the test have scaffolds count less than 100 and coverage more than 99%



DNBelab-D4 + DNBSEQ-E25 sequencing result

D4 cartridge	sample	DNB con. ng/μL	Total reads (M)	Q30(%)	Split rate (%)	Scaffolds count	Coverage (%)
Lane 1	Salmonella enterica-1	22.2	26.27	90.35	96.59	49	100
Lane 2	Salmonella enterica-2	25.5				53	99.59
Lane 3	Vibrio parahaemolyticus-1	19.2				66	100
Lane 4	Vibrio parahaemolyticus-2	21.2				67	99.81

DNBelab-D4 + DNBSEQ-G99 sequencing result

D4 cartridge	D4 Lane	sample	DNB con. ng/μL	Total reads (M)	Q30(%)	Split rate (%)	Scaffolds count	Coverage (%)
cartridge 1	Lane 1	Salmonella enterica-1	16.47	102.09	95.77	96.95	44	100
	Lane 2	Salmonella enterica-2	18.60				45	100
	Lane 3	Salmonella enterica-3	13.81				42	100
	Lane 4	Acinetobacter baumannii-1	14.42				89	100
cartridge 2	Lane 1	Vibrio parahaemolyticus-1	18.60				67	100
	Lane 2	Vibrio parahaemolyticus-2	16.53				63	100
	Lane 3	Vibrio parahaemolyticus-3	18.20				92	99.27
	Lane 4	Acinetobacter baumannii-1	13.81				91	100

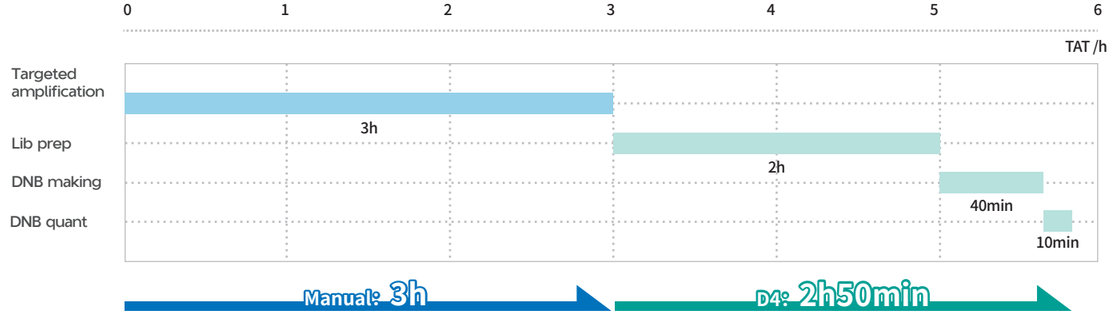
Case 2: Covid-19 Virus Genome Sequencing

Experiment Scheme

- **Sample:** mixture of virus RNA and human nucleic acid
- **Manual:** targeted amplification and purification
- **D4:** amplicon in, DNB out with DNB quant result
  Library prep： ATOPlex RNA Multiplex PCR Amplification Set V3.1,
  DNBelab-D4RS Fast PCR-FREE FS Library Prep Set V2.0
- **TAT:** manual: 3h, D4 2h50min
  (Including DNB making and DNB quantification)
- **Sequencing:** DNBSEQ-E25 SE100+10+10
- **Bioinformatics:** MGI metargetCOVID

Conclusions

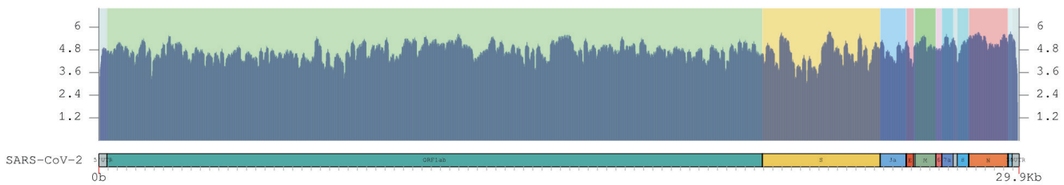
- DNBelab-D4 can obtain DNB with sufficient concentration (≥10ng/μL)
- DNBSEQ-E25 generated 28.57 M reads with high quality for analysis
- The mapping rate is more than 99% and 100X coverage is more than 99%.



DNBelab-D4 + DNBSEQ-E25 sequencing result

D4 cartridge	Sample	DNB con. ng/μL	Total reads (M)	Q30(%)	SplitRate (%)	Mapping rate (%)	1X Coverage (%)	30X Coverage (%)	100X Coverage (%)	Average depth
Lane 1	A	23.6	28.57	94.49	98.41	99.38	99.75	99.75	99.75	10258
Lane 2	B	16.7				99.46	99.75	99.75	99.73	9925
Lane 3	C	21.6				99.45	99.75	99.75	99.74	9925
Lane 4	D	24.4				99.35	99.75	99.75	99.75	9680

Histograms of coverage range



The X axis is position of the base in reference chromosome and Y axis is log10 value of reads depth

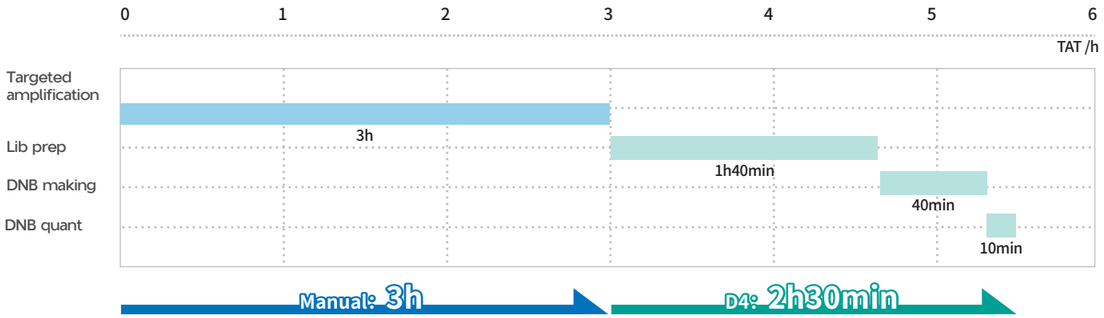
Case 3: Influenza Virus Genome Sequencing

Experiment Scheme

- **Sample:** mixture of virus RNA and human nucleic acid
- **Manual:** targeted amplification and purification
- **D4:** amplicon in, DNB out with DNB quant result
- **Library prep：** MGIEasy Respiratory Microorganisms Genome Amplification Kit , DNBelab-D4RS Fast PCR-FREE FS Library Prep Set V2.0
- **TAT**: manual: 3h, D4 2.5h(Including DNB making and DNB quantification)
- **Sequencing**: DNBSEQ-E25 PE100+10+10
DNBSEQ-G99 PE100+10+10
- **Bioinformatics**: MGI FluTrack

Conclusions

- DNBelab-D4 can obtain DNB with sufficient concentration (⩾10ng/μL)
- Both DNBSEQ-E25 and DNBSEQ-G99 can generate sufficient data amount with high quality for analysis
- The identification of influenza virus with type is correct. The mapping rate is more than 95% and completeness of assembly is more than 99%.



DNBelab-D4 + DNBSEQ-G99 sequencing result

D4 Lane	DNB con. ng/μL	Total reads(M)	Q30 (%)	Mapping rate(%)	Influenza identification	Influenza reads Pct(%)	Influenza type
Lane 1	26.2	101.2	95.73	97.66	Influenza A	93.14%	H1N1
Lane 2	21.6			97.70	Influenza A	93.27%	H1N1
Lane 3	25.8			97.63	Influenza A	92.94%	H1N1
Lane 4	28.2			97.60	Influenza A	93.27%	H1N1

DNBelab-D4 + DNBSEQ-E25 sequencing result

D4 Lane	DNB con. ng/μL	Total reads(M)	Q30 (%)	Mapping rate(%)	Influenza identification	Influenza reads Pct(%)	Influenza type
Lane 1	29.3	25.7	92.5	96.64	Influenza B	96.57	Victoria
Lane 2	30.9			97.33	Influenza B	96.63	Victoria
Lane 3	27.4			96.80	Influenza B	97.35	Victoria
Lane 4	27.9			96.54	Influenza B	96.86	Victoria

DNBelab-D4+DNBSEQ-E25 sequencing result

Gene	Genome segment	Q. Start (Influenza sequence)	Q. End (Influenza sequence)	S. Start (Influenza reference sequence)	S. End (Influenza reference sequence)	Alignment length	Segment length	Completeness of assembly
PB1	B-seg1	1	2364	4	2367	2364	2369	99.79%
PB2	B-seg2	1	2392	5	2396	2392	2396	99.83%
PA	B-seg3	1	2295	5	2299	2295	2305	99.57%
HA	B-seg4	1	1876	1	1882	1882	1882	100.00%
NP	B-seg5	1	1838	1	1838	1838	1844	99.67%
NA	B-seg6	1	1543	15	1557	1543	1557	99.10%
M	B-seg7	1	1189	1	1190	1190	1190	100.00%
NS	B-seg8	1	1096	6	1097	1096	1097	99.91%

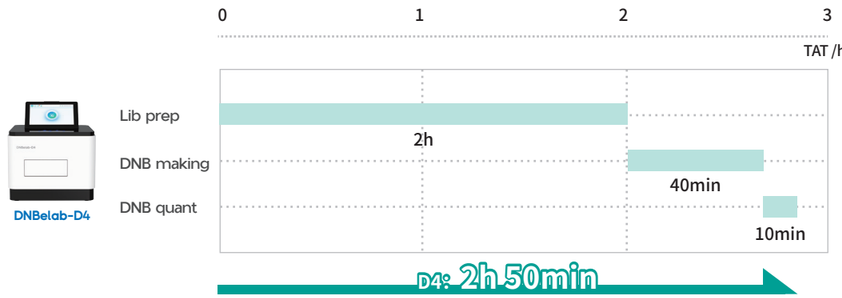
Case 4: Metagenomics Sequencing

Experiment Scheme

- **Sample:** human DNA mixed with microbial community DNA with different ratios
- **D4:** DNA in, DNB out with DNB quant result
- **Library prep Kit：** DNBelab-D4RS FS DNA Library Prep Set V2.0
- **D4 TAT:** 2h50min (Including DNB making and DNB quantification)
- **Sequencing:** DNBSEQ-G99 SE100+10+10
- **Bioinformatics:** PFI

Conclusions

- DNBelab-D4 can obtain DNB with sufficient concentration (⩾10ng/μL)
- DNBSEQ-G99 can generate sufficient data amount with high quality for analysis
- Pathogen identification results using PFI are consistent with the species in the standard sample.
- The relative abundance ranking of pathogen in samples with different ratios of microbial community DNA is consistent.



DNBelab-D4+DNBSEQ-G99 sequencing result

D4 lane	sample*	DNB con. (ng/μL)	Total reads (M)	Q30(%)	Split rate (%)
Lane 1	S1-5% Microbial community DNA	24.90	103.79	96.35	89.86%
Lane 2	S2-1% Microbial community DNA	21.28			
Lane 3	S3-2% Microbial community DNA	25.53			
Lane 4	S4-Microbial community DNA	21.27			

* The four lanes used the same microbial community DNA. The ratios of microbial community DNA in human DNA are different.

Relative abundance of microbes in each sample

	1% Microbial community DNA	2% Microbial community DNA	5% Microbial community DNA	100% Microbial community DNA
Salmonella enterica	16.84%	16.40%	15.61%	15.62%
Escherichia coli	13.98%	13.62%	13.17%	14.53%
Staphylococcus aureus	12.98%	12.81%	12.54%	13.48%
Bacillus subtilis	12.54%	12.58%	12.24%	13.17%
Pseudomonas aeruginosa	11.20%	10.92%	12.24%	12.18%
Enterococcus faecalis	11.02%	10.84%	10.99%	12.08%
Listeria monocytogenes	10.20%	10.05%	10.59%	10.51%
Limosilactobacillus fermentum	9.39%	9.21%	9.22%	9.66%
Saccharomyces cerevisiae	1.97%	1.79%	1.71%	1.77%
Cryptococcus neoformans	1.87%	1.77%	1.68%	1.68%