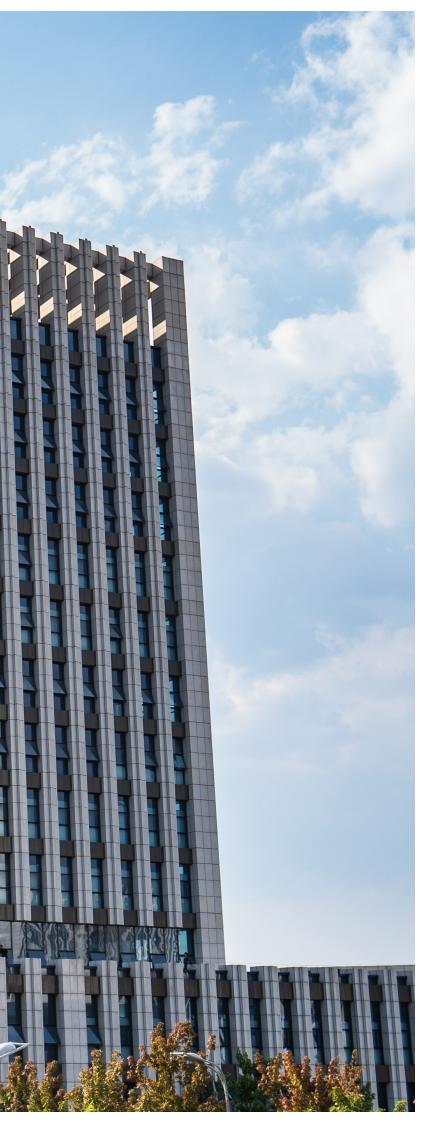




### RNA-seq Related Products for Illumina

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### 2012 2022

#### **About Vazyme**

Founded in 2012, Vazyme is now a leading biotechnology company in China, conducting research and development both in technology and products focusing on functional proteins, such as enzymes, antigens, antibodies, and polymer organic materials. Relying on an in-house generic technology platform, Vazyme has successively moved into various business areas ranging from biological research, in vitro diagnosis (IVD) to biopharmaceutical. Equipped with both independent technology development and terminal products manufacturing capabilities, Vazyme is now ready to power the biotechnology industry.





#### **Development of Core Technology**

Since its establishment, Vazyme has always adhered to the business philosophy—— R&D is the core. Through years of relentless efforts, Vazyme has achieved a lot in biomedical science. For instance, we have developed various kinds of biological preparations, covering but not limited to high-throughput sequencing library series, PCR, qPCR, molecular clone, reverse transcription and 8 sets of POCT diagnostic reagents used to detect heart and cerebral vessels, inflammation, sound child rearing, and gastric function, etc. Now Vazyme has expanded the customer portfolio to a wider range, including scientific research institutions, high-throughput sequencing service companies, molecular diagnostic reagent manufacturers, pharmaceutical companies, CRO companies, hospitals and other medical institutions.

#### **Innovation & Reservation of Talents**

Based on self-established core technologies, Vazyme has built an independent generic technology platform to meet the needs of large-scale research and development of products quickly and efficiently. Now we have over 200 kinds of genetic engineering recombinases, more than 1000 kinds of high-performance antigens, mAb and other critical materials. In addition, Vazyme owns above 500 terminal products that are widely applied in science research, high-throughput sequencing, IVD, pharmaceutical and vaccine research development, animal quarantine, etc.

Vazyme's powerful R&D strength is supported by a strong research and innovation team with over 400 multi-disciplinary experts majoring in molecular biology, enzymology, immunology, bioinformatics, organic chemistry and materials science, etc., more than half of which have a master's degree or above. We have been mentioned in CNS and sub-journals over 180 times and more than 1500 times in various other journals. The total citation of papers so far has reached to 10000.

#### Staying True to the Original Aspiration & Fulfill the Mission

On the way to deepen and broaden the entire product and technology chain, Vazyme stays true to the original aspiration —— "Science and Technology Make a Healthier Life", explores new methods for disease discovery, diagnosis, prevention and treatment, and provides quality products and professional services to creat value for clients.

In the past few years, Vazyme has actively engaged in the construction of public health programs, and played a vital role in fighting against African swine fever and COVID-19. **So far we have business cooperation with more than 300 IVD kit manufacturers by providing raw materials and premixes for over 800 million population covering more than 30 countries.** In the future, we will continue to contribute to the development of biosafety, and help mankind to overcome the threats posed by major infectious diseases, tumors, and autoimmune diseases!

**Va**ne™

UN864

## Product Catalogue

## Contents

#### **DNA Total Solution for Illumina**

Background	
RNA Quality Control	P01-04
RNA Enrichment	P05-11
RNA-Seq Library Preparation	P12-16
Adapters	P17-18
Purification and Size Selection	P19-21
Library Quality Control	P22-25

### Background

#### Brief Introduction

Transcriptome sequencing (RNA-seq) refers to the use of second generation high-throughput sequencing technology to complete and rapidly obtain almost all transcripts of a specific organ or tissue of a certain species under a certain state, mainly including mRNA and non-coding RNA (IncRNA, circRNA, small RNA). In a broad sense, transcriptome refers to the collection of all transcription products in a cell under certain physiological conditions, including mRNA, rRNA, tRNA and other non-coding RNA. While rRNA, tRNA and other non-coding RNA carry less genetic information, in the narrow sense transcriptome means the set of all mRNA.

Transcriptome study can study gene function and gene structure from an overall level, and reveal specific biological processes and molecular mechanisms in the process of disease occurrence. RNA-seq has been applied in many fields. For example, study on gene transcription level, such as gene expression, differentially expressed genes. Study on the structure of the transcript, such as variable splicing, gene fusion. And new transcript prediction.

#### Experimental Process

#### **RNA Enrichment**

The purpose of sequencing is to obtain more biological information. But the content of rRNA accounted for more than 80% of total RNA, can only provide very little information about the transcript. And the detection of too much rRNA will mask the expression richness of other genes. Therefore, rRNA is usually removed from RNA samples prior to sequencing. Different enrichment strategies can be selected according to experimental requirements or experimental technical conditions. First, when the information of mRNA needs to be obtained, the mRNA enrichment strategy can be selected. mRNA enrichment is based on the feature of eukaryotic mRNA with Ploy (A), and the combination of oligo (dT) magnetic beads and 3' Ploy (A) tail is used for enrichment. The second is rRNA removal, which can be selected when non-coding RNA information also needs to be obtained, or the incomplete RNA with RIN values less than 7, including FFPE samples.

#### **RNA-seq Library Preparation**

The general process of RNA library construction is as follows: firstly, enrich RNA, then interrupt mRNA under high temperature and high salt conditions, and add random primers for reverse transcription to synthesize the 1st strand cDNA. The 1st strand cDNA is unstable, and the 2nd strand should be synthesized immediately. Then the end need to be repaired, adding phosphate group to the 5' end and A tail to the 3' end, followed by adapter ligation, and finally PCR amplification.

The small RNA library can be constructed by the 3'/5' ends adapter ligation strategy. The 3' and 5' ends of small RNA are ligated to common adapters, respectively, and the sequencing libraries suitable for Illumina platform are finally obtained through reverse transcription, PCR amplification, PAGE gel or magnetic bead purification and other steps.

#### **Library Quality Control**

Quality control of library is very important before sequencing. The quality control process generally includes two aspects: concentration detection and library fragment distribution detection. After the library is constructed, the concentration of the library is determined first. There are two methods of library quantification: one is based on dsDNA fluorescent dyes, the other is qPCR-based quantification. Then the library fragment distribution can be detected using Aglient 2100 Bioanalyzer or an equivalent instrument. If the library insert fragment is too short, the reads containing the adapter will generally be filtered out (except for small RNA libraries), which will cause data waste. However, if the inserted fragment is too long, the efficiency of DNA clustering will be reduced, and the quality of sequencing will also be reduced.

#### RNA Quality Control

Product Name	Feature	Size	Cat No.#
Equalbit® RNA HS Assay Kit	- RNA concentration detection with Qubit	100/500 assays	EQ211-01/02
Equalbit® RNA BR Assay Kit	RNA concentration detection with Qubit	100/500 assays	EQ212-01/02

#### RNA Enrichment

Product Name	Feature	Size	Cat No.#
VAHTS® mRNA Capture Beads	mRNA enrichment	24/96 rxn	N401-01/02
Ribo-off <sup>®</sup> rRNA Depletion Kit (Human/Mouse/Rat)		24/96 rxn	N406-01/02
Ribo-off <sup>®</sup> rRNA Depletion Kit (Bacteria)	DAIA develotion	12/24 rxn	N407-01/02
Ribo-off <sup>®</sup> Globin & rRNA Depletion Kit (Human/Mouse/Rat)	rRNA depletion	24/96 rxn	N408-01/02
Ribo-off <sup>®</sup> rRNA Depletion Kit (Plant)		12/24 rxn	N409-01/02
VAHTS® RNA Clean Beads	RNA purification	5/40/450 ml	N412-01/02/03

#### RNA-seq Library Preparation

Product Name	Feature	Size	Cat No.#
VAHTS® Universal V8 RNA-seq Library Prep Kit for Illumina	RNA library construction	24/96 rxn	NR605-01/02
VAHTS® Small RNA Library Prep Kit for Illumina	Small RNA library construction	24/96 rxn	NR801-01/02

#### Adapters

Product Name	Feature	Size	Cat No.#
VAHTS® RNA Adapters Set 1/Set 2 for Illumina	Single index adapter for Illumina	10/40 µl each	N803/N804-01/02
VAHTS <sup>®</sup> RNA Adapters Set 3 - Set 6 for Illumina	Single index adapter for Illumina	20 µl each	N809-812
VAHTS <sup>®</sup> RNA Multiplex Oligos Set 1/Set 2 for Illumina	Dual index adapter for Illumina	192 rxn each	N323/N324
VAHTS <sup>®</sup> Small RNA Index Primer kit for Illumina	Small RNA index primer for Illumina	48 rxn each	N813-816

#### Purification and Size Selection

Product Name	Feature	Size	Cat No.#
VAHTS <sup>®</sup> DNA Clean Beads	Library purification or size selection	5/60/450 ml	N411-01/02/03

#### Library Quality Control

Product Name	Feature	Size	Cat No.#
VAHTS <sup>®</sup> Library Quantification Kit for Illumina	Library quantification	500 rxn each	NQ101-104
Equalbit® dsDNA HS Assay Kit	Library concentration detection with	100/500 assays	EQ111-01/02
Equalbit® 1 × dsDNA HS Assay Kit	Qubit	100/500 assays	EQ121-01/02



# RNA Quality Control

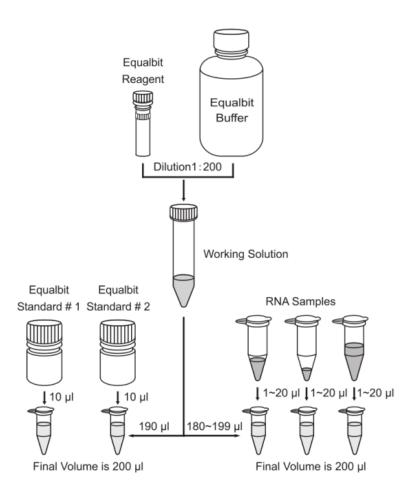
Product Name	Feature	Size	Cat No.#
Equalbit® RNA HS Assay Kit	DNA concentration detection with Oubit	100/500 assays	EQ211-01/02
Equalbit® RNA BR Assay Kit		100/500 assays	EQ212-01/02



#### **Equalbit RNA HS Assay Kit**

#### Description

The Equalbit RNA HS (High Sensitivity) Assay Kit is a simple, sensitive and accurate RNA fluorescence assay kit. The kit is highly selective for RNA and is not affected by dsDNA. It has excellent linearity for RNA samples in the range of 5 - 100 ng, and can accurately quantify total RNA, rRNA and mRNA samples at concentrations of 250 pg/µl to 100 ng/µl. It has excellent tolerance to some conventional pollutants, such as salt, free nucleotides, proteins, solvents, detergents, etc.



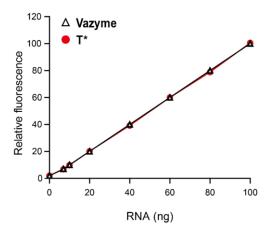
#### Features

#### 1/ Simple Operation

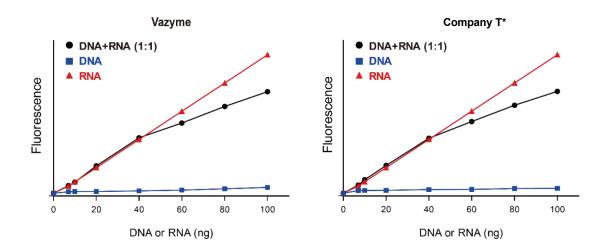
This product is easy to operate and can be carried out at room temperature. Before use, the fluorescence detection reagent is diluted into working solution with buffer solution, and then the RNA sample to be tested is added to the Qubit Fluorescence Analyzer for detection.

#### 2/ Precise Quantification

All the RNA samples had a good linear relationship in the range of 5 - 100 ng. The values of low concentration (5 - 10 ng) and high concentration were all linear.



The detection effects of EQ211 product and T\* company similar product are consistent, both have good specificity for RNA, and the binding ability to dsDNA is extremely weak.





The EQ211 product has same tolerance to different impurities as similar products of T\* company when the sample to be tested contains contaminants.

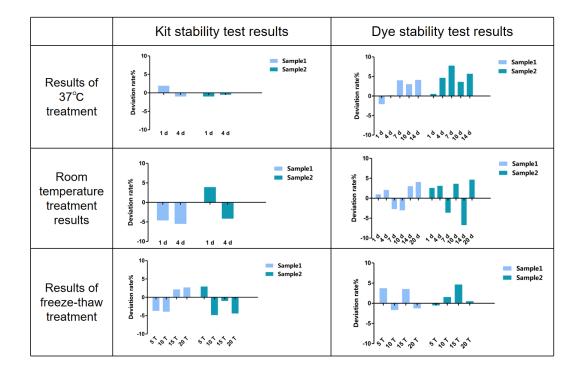
Table 1. Impurity tolerance test

Contaminant	Final concentration in	Result
Contaminant	the assay	(Compared with similar products from T* Company)
NaCl	10 mM	$\sqrt{}$
NH <sub>4</sub> AC	10 mM	$\sqrt{}$
Ethanol	1%	V
Phenol	0.1%	V
Chloroform	0.2%	$\sqrt{}$
Triton X-100	0.001%	V
dNTP	100 μΜ	$\sqrt{}$
BSA	20 μg/ml	V
ssDNA	1×*	V
Oligos	1×*	V
dsDNA	1×*	√

<sup>\* 1×</sup> indicates the same concentration as RNA.

#### 3/ High Stability

EQ211 has good stability between batches. Accurate test results can be obtained by placing the whole kit at 37°C for 4 days, room temperature for 4 days, and repeated freezing and thawing for 20 times. The dye can be placed at 37°C for 14 days, room temperature for 20 days and repeated freezing and thawing for 20 times, respectively, to obtain accurate test results.





## RNA Enrichment

Product Name	Feature	Size	Cat No.#
VAHTS® mRNA Capture Beads	mRNA enrichment	24/96 rxn	N401-01/02
Ribo-off® rRNA Depletion Kit (Human/Mouse/Rat)		24/96 rxn	N406-01/02
Ribo-off® rRNA Depletion Kit (Bacteria)	#DNA doplation	12/24 rxn	N407-01/02
Ribo-off® Globin & rRNA Depletion Kit (Human/Mouse/Rat)	rRNA depletion	24/96 rxn	N408-01/02
Ribo-off® rRNA Depletion Kit (Plant)		12/24 rxn	N409-01/02
VAHTS <sup>®</sup> RNA Clean Beads	RNA purification	5/40/450 ml	N412-01/02/03

#### **VAHTS mRNA Capture Beads**

#### Description

mRNA Capture Beads is 1 µm paramagnetic microspheres coupled with Oligo (dT) suitable for poly (A)+ RNA separation from purified total RNA.

#### Working Principle



#### Features

#### 1/ Simple Operation

The magnetic separation technique can isolate the intact mRNA from the small volume of samples and avoid the step of mRNA precipitation.

#### 2/ Rapid

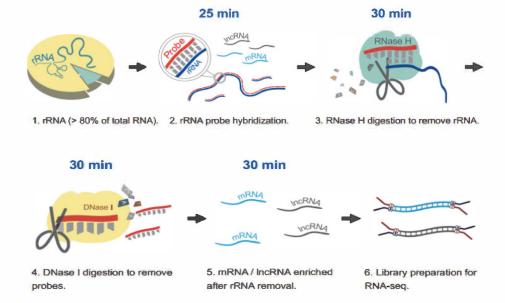
The whole operation process can be completed within 1 h.

#### Ribo-off rRNA Depletion Kit (Human/Mouse/Rat)

#### Description

Ribo-off rRNA Depletion Kit (Human/Mouse/Rat) is a kit for removing rRNA from total RNA (including cytoplasmic 28S, 18S, 5S rRNA and mitochondrial 12S, 5.8S rRNA), leaving other mRNA and non-coding RNA. The kit is applicable to both complete and partially degraded RNA samples (e.g. FFPE RNA). And the resulting rRNA-depleted RNA can be analyzed for both mRNA and non-coding RNA such as IncRNA.

#### Working Principle



#### Features

#### 1/Rapid

Complete rRNA removal within 2 h.

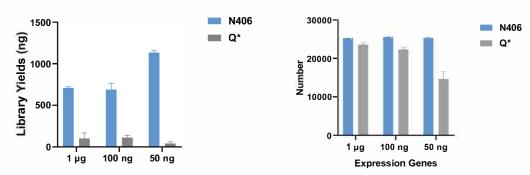
#### 2/ Wide Compatibility

Extensive species compatibility.

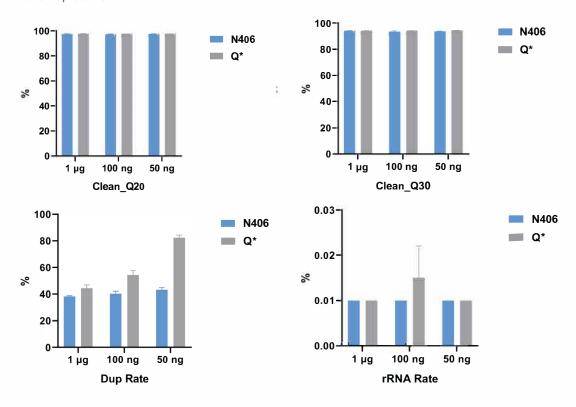
Ното	Mus	Rattus	Bos	Canis lupus	Equus	Gallus	Macaca	Sus
sapiens	musculus	norvegicus	taurus	familiaris	caballus	gallus	mulatta	scrofa
Human	Mouse	Rat	Cow	Dog	Horse	Chicken	Monkey	Pig

#### 3/ High Efficiency

Ribo-off rRNA Depletion Kit (Human/Mouse/Rat) can be efficiently compatible with different input quantities.

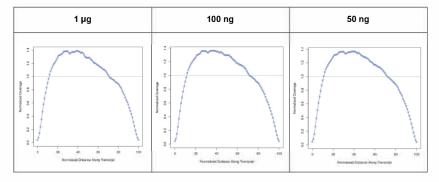


The transcriptome library was constructed with N406 and VAHTS Universal V6 RNA-seq Library Prep Kit for Illumina (Vazyme #NR604), with good data quality and high rRNA removal efficiency (>99.9%). The dup rate of rRNA removal products were significantly better than Q\* company rRNA removal products.



#### 4/ High Data Uniformity

Use N406 with VAHTS Universal V6 RNAseq Library Prep Kit for Illumina (Vazyme #NR604) to construct a transcriptome library with high uniformity.



#### Ribo-off Globin & rRNA Depletion Kit (Human/Mouse/Rat)

#### Description

This is a kit for removing globin mRNA and rRNA from total RNA in blood samples. This kit is compatible with 0.01 - 1 µg blood total RNA. The total RNA sample undergoes probe hybridization, RNase H digestion, DNase I digestion, etc. Finally, globin mRNA and rRNA (including cytoplasmic rRNA and mitochondrial rRNA) are removed from total RNA, leaving other mRNA and non-coding RNA, which can be used for the analysis of non-coding RNA such as IncRNA.

#### Working Principle

1. Globin mRNA and rRNA hybridize with probe (Probe hybridization)



#### Features

#### 1/ Wide Compatibility

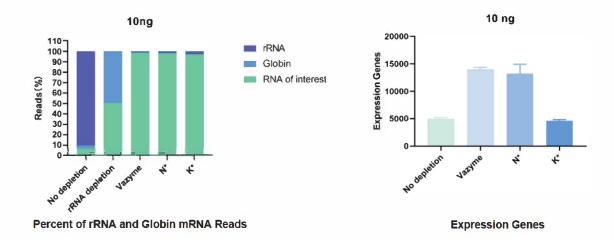
Compatible with different species, different starting quantities and different quality samples. And both globin mRNA and rRNA can be removed.

#### 2/ High Library Yield

Under different species and different inputs, Vazyme N408 can be efficiently compatible with library modules, ensuring good library output.

#### 3/ High Efficiency

Vazyme N408 could effectively remove globin mRNA and rRNA in different samples with different initial inputs (0.01 -  $1\mu g$ ), different species and different quality. And the kit was paired with VAHTS Universal V8 RNA-seq Library Prep Kit for Illumina (Vazyme #NR605) for stranded transcriptome library construction. In the index of gene detection, it was consistent with that of N\* similar products to ensure a rich number of gene detection.



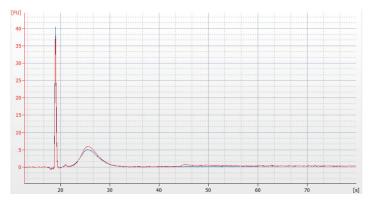
#### **VAHTS RNA Clean Beads**

#### Description

The Vazyme VAHTS RNA Clean Beads utilizes on SPRI (Solid Phase Reverse Immobilization) and is applicable for RNA purification. This kit selectively binds RNA to the beads and efficiently removes all proteins, salt ions, and other impurities. The usage of VAHTS RNA Clean Beads is the same as the Agencourt RNAClean XP Beads, which is widely used currently. The cost-effective VAHTS RNA Clean Beads serves as a seamless alternative for Agencourt RNAClean XP Beads.

#### Applications

Applicable for RNA purification from in vitro reaction mixtures, such as RNA library preparation. Not applicable for direct RNA purification from cells or tissues.



- Agencourt "RNAClean" XP
- VAHTS RNA Clean Beads

After 100 ng UHRR (Universal Human Reference RNA) was segmented at 85°C for 5 min, it was purified with 2.2 X VAHTS RNA Clean Beads and Agencourt RNAClean XP, and then detected with Agilent 2100 Bioanalyzer.

#### Comparison of Recovery Rates Between Vazyme and Beckman RNA Clean Beads

Fragment Size (nt)	100	200	300	400	500	600	800	1000	1500	2000
Input Concentration (ng/µl)	171.41	169.22	195.6	97.79	46.38	37.15	38.79	38.1	45.46	46.95
Xp Recovery Concentration (ng/μl)	143.92	142.54	160.37	90.09	37.18	28.71	35.91	34.65	39.36	41.2
$\begin{array}{c} Vazyme \ Recovery \\ Concentration \ (ng/\mu l) \end{array}$	137.94	138.28	155.15	92.62	39.25	31.39	38.33	33.61	39.71	39.71
Xp Recovery Rate	83.96%	84.23%	81.99%	92.12%	80.16%	77.29%	92.59%	90.94%	86.59%	87.75%
Vazyme Recovery Rate (%)	80.47%	81.72%	79.32%	94.71%	84.62%	84.50%	98.81%	88.23%	87.35%	84.57%
Vazyme/Xp	95.80%	97.00%	96.70%	102.80%	105.60%	109.30%	106.70%	97.00%	100.90%	96.40%

# RNA-seq Library Preparation Preparation

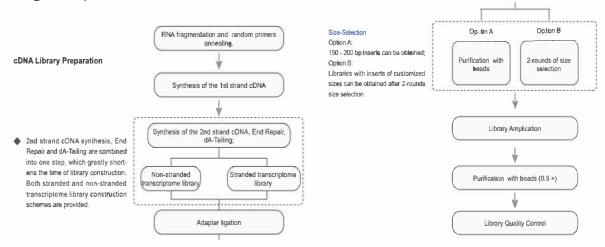
Product Name	Feature	Size	Cat No.#
VAHTS® Universal V8 RNA-seq Library Prep Kit for Illumina	RNA library construction	24/96 rxn	NR605-01/02
VAHTS® Small RNA Library Prep Kit for Illumina	Small RNA library construction	24/96 rxn	NR801-01/02

#### VAHTS Universal V8 RNA-seq Library Prep Kit for Illumina

#### Description

VAHTS Universal V8 RNA-seq Library Prep Kit for Illumina is specially designed for the preparation of transcriptome libraries for Illumina platform. The kit is perfectly compatible with all samples of total RNA, with the minimum initial input as low as 10 ng. The kit is universal and suitable for RNA library construction of RNA that have been obtained by Poly (A)-based mRNA enrichment or rRNA depletion. The kit contains two types of cDNA 2nd strand synthesis buffer, which can be chosen for library construction for non-stranded or stranded transcriptome analysis. This kit combines 2nd strand cDNA synthesis, end-repair and dA-tailing into one step, with no need of purification, which greatly simplifies the process of library construction and shortens the operation time.

#### Working Principle



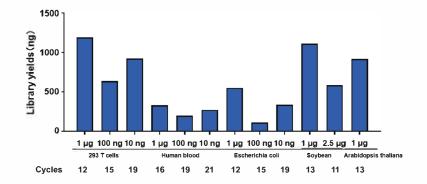
#### Features

#### 1/Rapid

The construction time of transcriptome library can be reduced to 3 h by the minimal operation process.

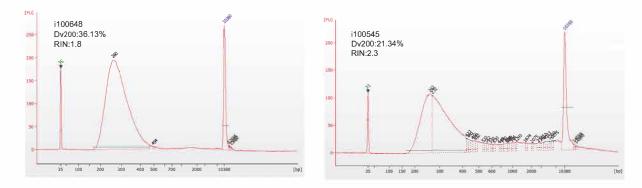
#### 2/ Extensive Species Compatibility

The stranded transcriptome library was constructed according to the library construction process of NR605. The results showed that libraries can be successfully constructed for different samples, and the yield is normal.



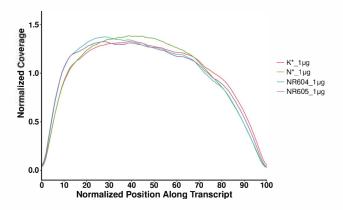
#### 3/ High Success Rate

The stranded transcriptome library was constructed according to the library construction process of NR605. The results showed that NR605 can be successfully used to build library for FFPE samples of different quality.



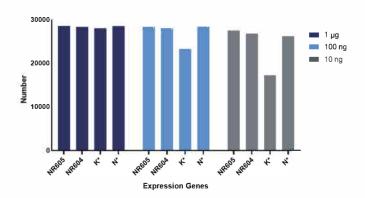
#### 4/ High Data Uniformity

Sequencing results showed that the libraries prepared by different transcriptome library kits were highly consistent in the uniformity of sequencing.



#### 5/ More Abundant Genes were Detected

The sequencing results showed that for the same sequencing quantity, the libraries constructed by using different transcriptome library preparation products could obtain a relatively rich number of genes detected. However, for low initial input, NR605 had more abundant gene detection numbers.

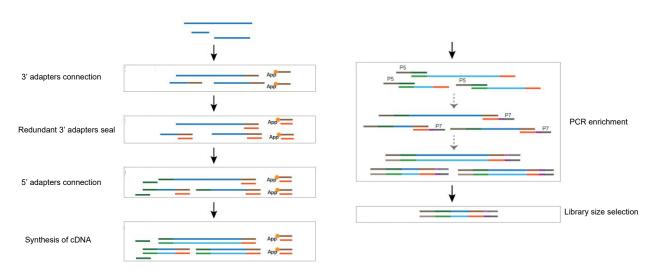


#### **VAHTS Small RNA Library Prep Kit for Illumina**

#### Description

VAHTS Small RNA Library Prep Kit for Illumina is a small RNA library construction Kit developed for Illumina high-throughput sequencing platform. This kit is suitable for a wide range of template types, animal and plant total RNA, as well as isolated and purified small RNA can be used as the starting template. Common adapters are added to the 3' and 5' ends of small RNA, respectively. After reverse transcription, PCR amplification, and purification with PAGE gel or magnetic beads, the Illumina sequencing library is finally obtained.

#### Working Principle



#### **Features**

#### 1/ Wide Compatibility

This kit is suitable for a wide range of template types, animal and plant total RNA, as well as isolated and purified small RNA can be used as the starting template.

The initial input is as low as 100 ng total RNA.

#### 2/ High Abundance of Library

The basic data indicators showed that the small RNA library constructed by VAHTS Small RNA Library Prep Kit for Illumina had a low contamination rate and a high proportion of effective libraries. The data comparison showed a high proportion of miRNA and a variety of miRNA and piRNA.

Sample _name	High_ quality	3'adapter_null or insert_null	5'adapter_ contaminants	Smaller_tags (less than 18nt)	Clean_reads _rate	mapping _rate	Known miRNA species	New miRNA species	piRNA species	miRNA rate
V1-1 µg	99.73%	1.57%	0.13%	7.87%	90.42%	88.20%	795	40	424	75.68%
V2-1 μg	99.72%	2.55%	0.10%	6.56%	90.81%	88.92%	818	52	423	75.99%
N-1 µg	99.83%	4.09%	0.14%	9.25%	86.47%	70.22%	760	27	376	56.50%
I-1 μg	99.74%	3.29%	0.41%	15.96%	80.26%	84.90%	829	70	388	68.21%
V1-100 ng	99.72%	6.53%	0.24%	8.72%	84.52%	86.67%	792	69	358	70.08%
V2-100 ng	99.75%	7.01%	0.27%	10.47%	82.23%	88.07%	729	74	375	70.27%
N-100 ng	99.77%	8.33%	0.23%	8.17%	83.15%	72.33%	723	59	385	51.12%
I-100 ng	99.71%	13.13%	2.04%	17.39%	67.29%	84.75%	764	50	320	67.69%

Note: HeLa cell total RNA was used as the starting template, and the inputs were 1  $\mu$ g and 100 ng, respectively. Among them, V1 and V2 were two parallel and repeated small RNA libraries constructed with NR801, N represents the library constructed with the small RNA library construction kit of company N, and I represents the library constructed with the small RNA library construction kit of company I.

#### 3/ Reliable Quality

The inserted fragments were distributed at 22 and 23 nt. The inserted fragment of small RNA library was normal in length.

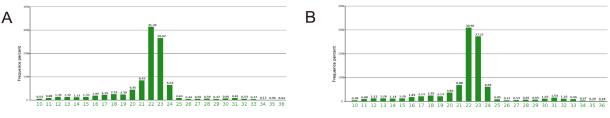


Fig A. 1 µg initial input small RNA libraries.

Fig B. 100 ng initial input small RNA libraries.

Small RNA annotation results showed that the proportion of miRNA was very high. The statistical results of the preference of the first and other bases showed that the first base had a strong preference for U, but repels G, and the second to fourth bases lack U. The VAHTS Small RNA Library Prep Kit for Illumina was used to construct small RNA libraries with reliable data quality.

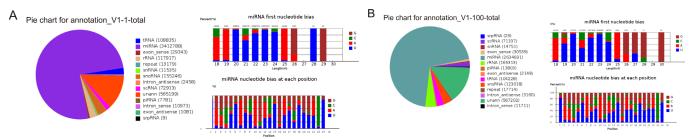


Fig A. 1 µg initial input small RNA libraries.

Fig B. 100 ng initial input small RNA libraries.

#### 4/ Reproducibility

The repetition correlation of libraries with the same initial input was greater than 0.99, and the correlation between libraries with different initial inputs was greater than 0.98, indicating that the small RNA library constructed using VAHTS Small RNA Library Prep Kit for Illumina had very good reproducibility.

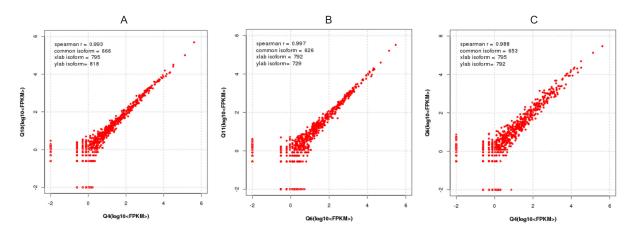


Fig A. Correlation between two 1 μg initial input small RNA libraries. Fig B. Correlation between two 100 μg initial input small RNA libraries. Fig C. Correlation between 1 μg and 100 ng initial input small RNA library.



## Adapters Adapters

Product Name	Feature	Size	Cat No.#	
VAHTS® RNA Adapters Set 1/Set 2 for Illumina	Single index adapter for Illumina	10/40 µl each	N803/N804-01/02	
VAHTS® RNA Adapters Set 3 - Set 6 for Illumina	Single index adapter for indiffina	20 µl each	N809-812	
VAHTS® RNA Multiplex Oligos Set 1/Set 2 for Illumina	Dual index adapter for Illumina	192 rxn each	N323/N324	
VAHTS® Small RNA Index Primer kit for Illumina	Small RNA index primer for Illumina	x primer for Illumina 48 rxn each		

#### **Adapters**

#### Description

- VAHTS RNA Adapters Set 1 6 for Illumina (Vazyme #N803/N804 or #N809/N810/N811/N812)
  - o N803/804: up to 24 kinds of single-ended 6-bp Indexed Adapters, 12 kinds/each set
  - o N809-812: up to 96 kinds of single-ended 8-bp Indexed Adapters, 24 kinds/each set



- VAHTS Multiplex Oligoes Set 4/5 for Illumina (Vazyme #N323/N324)
  - N323/N324: universal adapter+8 kinds of i5 primer+12 kinds of i7 primer/each set, allowing the construction of up to 384 different libraries.



- VAHTS Small Index Primer Kit for Illumina (Vazyme #N813/N814/N815/N816)
  - o N813-816: Contains 1 kind of universal primer and 48 kinds of various small RNA Index Primer



### Purification

Purification and Size Selection

# and Size Selection

Product Name	Feature	Size	Cat No.#
VAHTS® DNA Clean Beads	Library purification or size selection	5/60/450 ml	N411-01/02/03

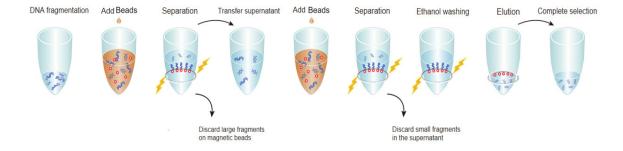
#### **VAHTS DNA Clean Beads**

#### Description

VAHTS DNA Clean Beads are based on the principle of SPRI (Solid Phase Reverse Immobilization), and are suitable for DNA purification and size selection for the high-throughput sequencing libraries construction. VAHTS DNA Clean Beads are compatible with DNA and RNA library building kits of various brands and library building processes reported in the literature. They are used in exactly the same way as AMPure XP Beads which are widely used at present. The production and size distribution of the library are consistent with the height of AMPure XP Beads. Therefore, AMPure XP Beads can be seamlessly replaced to effectively reduce the cost of library construction.

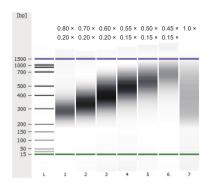
#### Working Principle

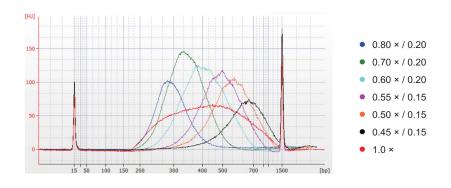
Through two steps of magnetic bead purification to achieve accurate separation effect.



Using TruePrep DNA Library Prep Kit V2 for Illumina (Vazyme #TD501) to construct DNA library. Library initial size is 200 - 1500 bp, using VAHTS DNA Clean Beads according to the following table to separation of PCR products.

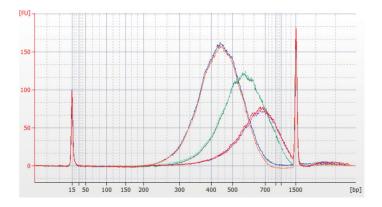
Volume ratio in the 1st round (Beads : DNA)	0.80 ×	0.70 ×	0.60 ×	0.55 ×	0.50 ×	0.45 ×	1.0 ×
Volume ratio in the 2nd round (Beads : DNA)	0.20 ×	0.20 ×	0.20 ×	0.15 ×	0.15 ×	0.15 ×	
Average library length (bp)	300	350	400	500	600	700	200-1500





#### ■ DNA Library Construction, Compared with AMPure XP Beads

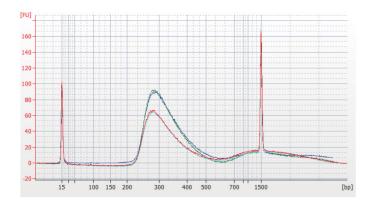
Using TruePrep DNA Library Prep Kit V2 for Illumina (Vazyme #TD501) to construct DNA Library with 50 ng the human DNA template. Using VAHTS DNA Clean Beads and AMPure XP Beads to select the average length of about 470, 570, 670 bp library in the same conditions (corresponding insert size is 350, 450, 550 bp). Using the Agilent 2100 Bioanalyzer to analyze.



- VAHTS<sup>™</sup> DNA Clean Beads 470 I
- AMPure XP Beads 470 bp
- VAHTS™ DNA Clean Beads 570 I
- AMPure XP Beads 570 bp
- VAHTS™ DNA Clean Beads 670 I
- AMPure XP Beads 670 bp

#### DNA Library Construction, Compared with AMPure XP Beads

Using TruSeq RNA Sample Prep Kit V2 (Illumina #RS-122-2001) to construct RNA Library with 1 µg or 100 ng Universal Human Reference RNA (UHRR) template. Using VAHTS DNA Clean Beads and AMPure XP Beads to select the average length of about 280 bp library in the same conditions (corresponding insert size is 160 bp). Using the Agilent 2100 Bioanalyzer to analyze.



- VAHTS<sup>™</sup> DNA Clean Beads 1 µg
- AMPure XP Beads 1 μg
- VAHTS™ DNA Clean Beads 100 ng
- AMPure XP Beads 100 ng

# Library Quality Control

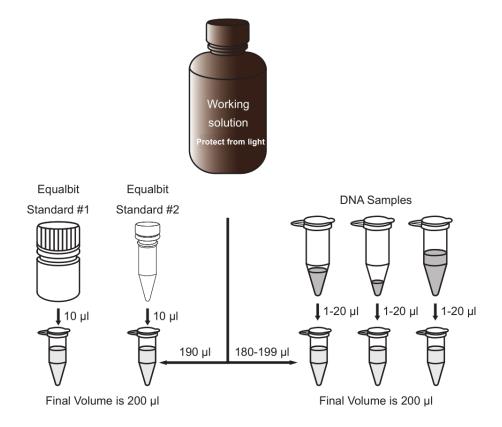
Product Name	Feature	Size	Cat No.#	
VAHTS <sup>®</sup> Library Quantification Kit for Illumina	Library quantification	500 rxn each	NQ101-104	
Equalbit <sup>®</sup> dsDNA HS Assay Kit	Library concentration detection with	100/500 assays	EQ111-01/02	
Equalbit <sup>®</sup> 1 × dsDNA HS Assay Kit	Qubit	100/500 assays	EQ121-01/02	

#### Equalbit 1 × dsDNA HS Assay Kit

#### Description

Equalbit 1 × dsDNA HS (High Sensitivity) Assay Kit is a simple, sensitive and accurate fluorescence quantitative assay kit for double stranded DNA (dsDNA). Equalbit 1 × dsDNA HS Assay Kit includes premixed working solution (containing fluorescent dye) and dsDNA standard. The kit has excellent linear relationship with dsDNA samples in the range of 0.2 - 100 ng, and can accurately quantify samples from 10 pg/µl to 100 ng/µl. For some conventional contaminants, such as RNA, salt, free nucleotides, proteins, solvents, detergents and so on have excellent tolerance.

#### Working Principle



#### Features

#### 1/ Simple Operation

During the experimental operation, there is no need to prepare working solution. The premix is directly divided into 0.5 ml tubes to add standard or test samples, and the data can be read using Qubit Fluorometer 2.0, 3.0, 4.0.

#### 2/ Simple Operation

For 12 dsDNA samples with different concentrations, Vazyme #EQ121 and similar products of T\* company were used for linear determination, and the fluorescence value was read by Qubit fluorometer 3.0. The results showed that the total amount of dsDNA samples has a good linear relationship in the range of 0.2 - 100 ng.

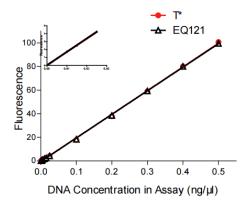


Fig 1. Comparison of 0.2-100 ng linear relationship

The results showed that the deviation rate between Vazyme #EQ121 and similar products of T\* company (difference between Vazyme #EQ121 and similar products of T\* company) was within 10% near the critical test point.

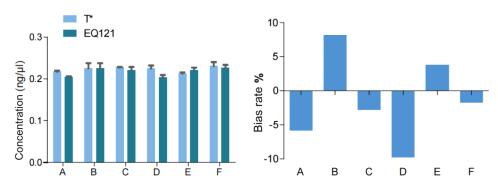


Fig 2. Plot of results for different operators testing low concentration as well as bias rate

#### 3/ Exceptional Specificity

The results showed that Equalbit 1 × dsDNA HS Assay Kit could specifically bind dsDNA, and even in the presence of RNA, it could still accurately quantify dsDNA, and its performance was comparable to that of similar products of T\* company.

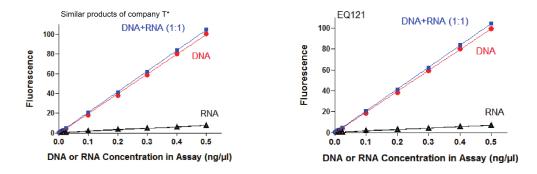


Fig 3. dsDNA specificity detection diagram of EQ121 and similar products of T\* company.

#### 4/ Dyes Bind Quickly

The results showed that the deviation between the test results of the two kit after adding samples for 5 min and 1 min was within 10%, indicating that the binding speed of the two kit to dsDNA samples was the same, and saturation could be achieved within 2 min.

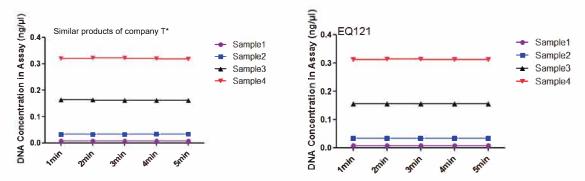


Fig 4. Binding speed of EQ121 with dsDNA dye of similar products of T\* company



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