sage science

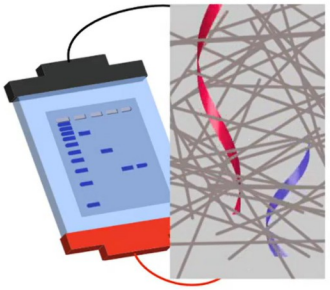
Pippin Pulse

脉冲场电泳系统

环亚生物科技有限公司（APG BIO LTD）

sage science

美国Sage Science公司位于美国马萨诸塞州Beverly市， 专注于核酸领域的样品制备。自2010年成立以来，核心产品Pippin系列全自动DNA片段回收仪因其优秀的性能表现， 得到全球超过3000家客户的认可，国内装机数量400台左右。超过2万篇文献中应用，包括众多Nature、Science、Cell系列的文章已成为DNA片段筛选回收领域的行业金标准。

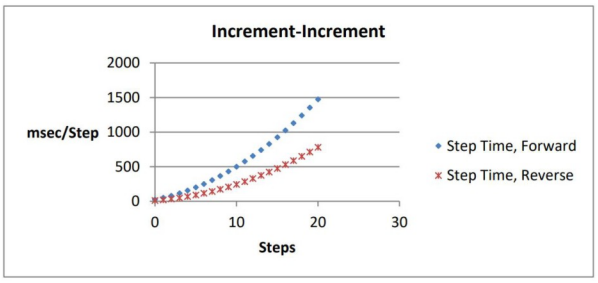


为什么需要脉冲场电泳?

普通琼脂糖凝胶电泳是利用分子筛的原理，分离不同大小的DNA片段。而当DNA分子大于10kb时，DNA分子的直径远远大于凝胶孔径，凝胶的分子筛就无法起到分离的作用。此时，DNA分子的迁移速率与其分子量大小不再呈现简单的相关性，因而导致无法通过观测大分子DNA的迁移速率来判断其分子量大小。

脉冲场电泳原理

通过快速地切换电场方向，有效地区别分离大分子DNA。脉冲场电泳（PFGE，Pulsed Field Gel Electrophoresis）是一种有效分离大分子DNA的方法。通过不断改变电场方向，驱使DNA在凝胶内前向和后向来回运动，小分子DNA片段改变方向的速率要比大分子DNA片段快。

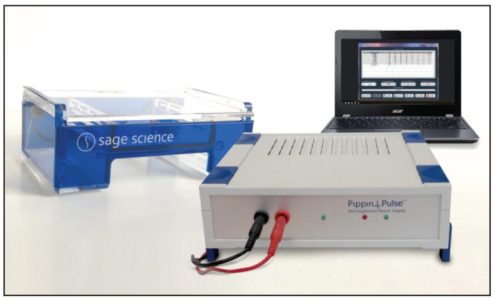


sage science

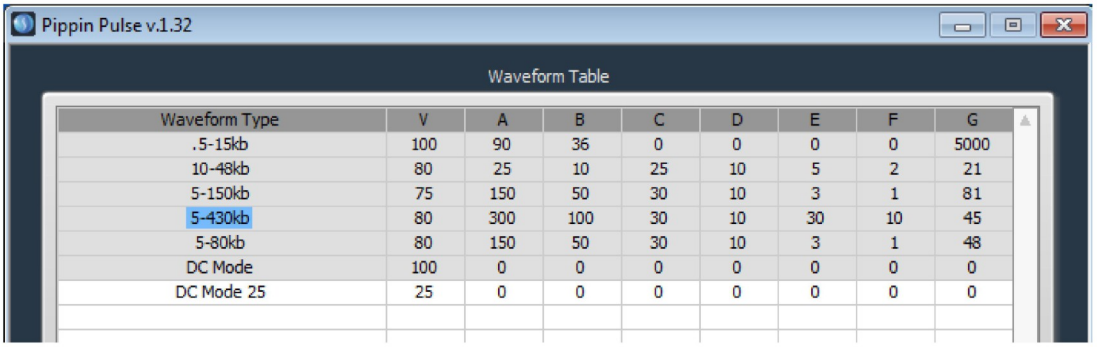
Pippin Pulse脉冲场电泳系统

特点：

- 大片段DNA质控，有效分离 0.5kb-430kb；
- 预设脉冲场程序，开机即用；
- 支持自定义程序；
- 稳定耐用，加固的大直径铂电极，寿命更长
- 试剂耗材开放，成本更低



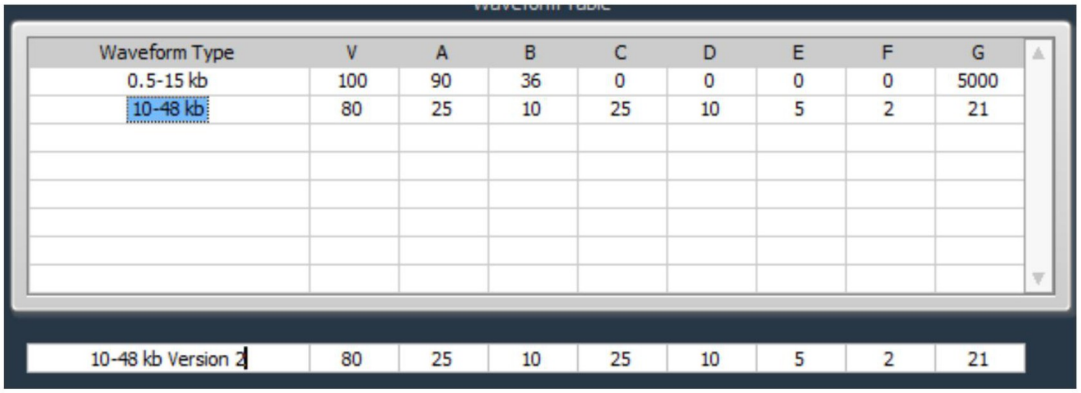
预设的脉冲场程序



Waveform Type	V	A	B	C	D	E	F	G
.5-15kb	100	90	36	0	0	0	0	5000
10-48kb	80	25	10	25	10	5	2	21
5-150kb	75	150	50	30	10	3	1	81
5-430kb	80	300	100	30	10	30	10	45
5-80kb	80	150	50	30	10	3	1	48
DC Mode	100	0	0	0	0	0	0	0
DC Mode 25	25	0	0	0	0	0	0	0

软件自带6种预设程序

支持自定义程序

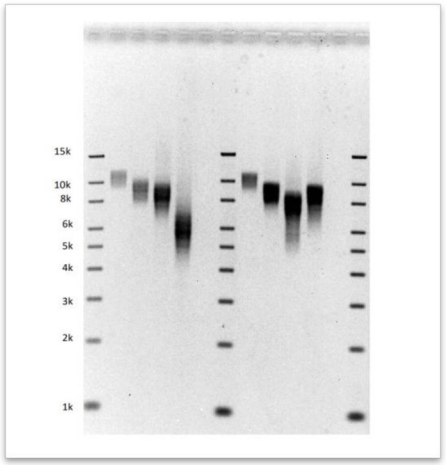


Waveform Type	V	A	B	C	D	E	F	G
0.5-15 kb	100	90	36	0	0	0	0	5000
10-48 kb	80	25	10	25	10	5	2	21
10-48 kb Version 2	80	25	10	25	10	5	2	21

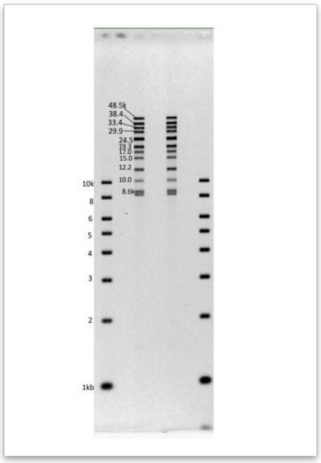
V.电压=电泳电压，25-175 V之间。
A.运行开始时的正向时间，1–65535毫秒。
B.运行开始时的反向时间，1–65535毫秒。
C.每一步增加到A的增量，0–255毫秒。
D.每一步增加到B的增量，0–255毫秒。
E.每一步增加到C的增量，0–255毫秒。
F.每一步增加到D的增量，0–255毫秒。
G.每个周期的步数，1–65535。

sage science

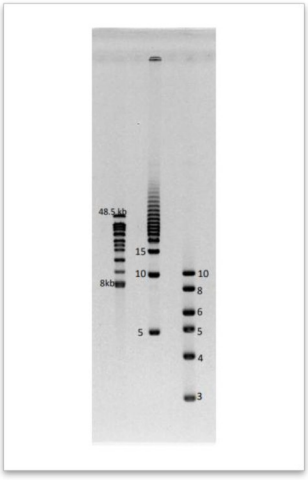
脉冲场电泳效果



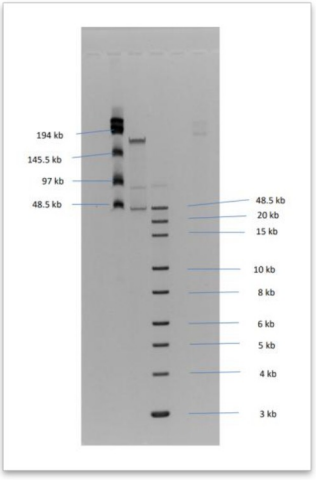
程序：5-15 kb
运行时间：8 hours
电压：100 V
缓冲液：0.5×KBB
琼脂糖胶：0.75% Lonza SeaKem GOLD, 110ml
Ladder: Biorad EZ Load 1kb Molecular Ruler # 170-8355



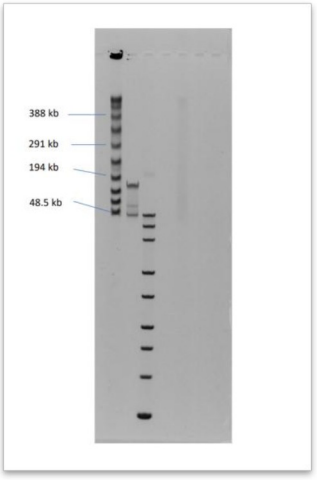
程序：10-48 kb
运行时间：9 hours
电压：80 V
缓冲液：0.5×KBB
琼脂糖胶：0.75% Lonza SeaKem GOLD, 110ml
Ladder: NEB Quick-load 1kb ladder #N0468S; Biorad CHEF DNA Size Standards 8-48 kb #170-3707



程序：5-80 kb
运行时间：14 hours
电压：80 V
缓冲液：0.5×TBE
琼脂糖胶：1% Lonza SeaKem GOLD, 110ml
Ladder: Biorad CHEF DNA Size Standards 8-48 kb #170-3707; Biorad DNA Size Standard 5 Kb Ladder Catalog # 170-3624



程序：5-150 kb
运行时间：16 hours
电压：75 V
缓冲液：0.5×KBB
琼脂糖胶：0.75% Lonza SeaKem GOLD, 110ml
Ladder: NEB Lambda PFG Ladder # N0341S; NEB 1kb Extend DNA Ladder #N3239



程序：5-430 kb
运行时间：16 hours
电压：75 V
缓冲液：0.5×KBB
琼脂糖胶：0.75% Lonza SeaKem GOLD, 110ml
Ladder: NEB Lambda PFG Ladder # N0341S; Biorad CHEF DNA Size Standards 8-48 kb #170-3707

sage science

顶级期刊

nature

Explore content ∨ About the journal ∨ Publish with us ∨

nature › articles › article

Article | Open Access | Published: 28 April 2021

Towards complete and error-free genome assemblies of all vertebrate species

Isolation of high-molecular-weight DNA

Agarose plug DNA isolation. For tissue, HMW DNA was extracted using the Bionano animal tissue DNA isolation fibrous tissue protocol (cat no. RE-013-10; document number 30071), according to the manufacturer's guidelines. A total of 25–30 mg was fixed in 2% formaldehyde and homogenized using the Qiagen TissueRuptor or manual tissue disruption. For nucleated blood, 27–54 µl was used with an adapted protocol (Bionano, personal communication) of the Bionano Prep Blood and Cell Culture DNA Isolation Kit (cat no. RE-130-10). Lysates were embedded into agarose plugs and treated with Proteinase K and RNase A. Plugs were then purified by drop dialysis with 1× TE. DNA quality was assessed using pulse field gel electrophoresis (PFGE) (Pippin Pulse, SAGE Science, Beverly, MA) or the Femto Pulse instrument (Agilent). PFGE revealed that we isolated ultra-high-molecular-weight DNA between ~100 and ~500 kb long.

Nature

Resource

Cell Reports

A chromosome-level reference genome and pangenome for barn swallow population genomics

METHOD DETAILS

DNA extraction

HMW (High Molecular Weight) DNA was extracted from the muscle tissue of the samples female barn swallow with the Bionano animal tissue DNA isolation fibrous tissue protocol (cat# RE-013-10; document number 30071). Approximately 55 mg of frozen muscle tissue was fixed in formaldehyde (2%) and homogenised with the Qiagen TissueRuptor. The lysate was included in agarose plugs, which were then treated with Proteinase K and RNase A. The DNA was recovered and purified from the plugs through a drop dialysis with 1x TE. Pulsed Field Gel Electrophoresis (PFGE; Pippin Pulse, SAGE Science, Beverly, MA) and Qubit were used for DNA quality control. According to the PFGE run, a large fraction of the isolated DNA was >250kbp.

Article

Cell Metabolism

Fatty acid overproduction by gut commensal microbiota exacerbates obesity

Graphical abstract Authors

Whole genome sequencing of microbes

E. coli TE-17 and *F. intestini* AJ110941 were cultivated as described above. The culture media was centrifuged, the cell pellet was washed once with PBS, and then stored at −80˚C until the DNA extraction. Bacterial genomic DNA was extracted as described above with slight modifications. Briefly, lysozyme and proteinase K were used for bacterial cell lysis. DNA quality was assessed using a spectrophotometer (DS-11, DeNovix) and Qubit fluorometer (Q33216, Thermo Fisher Scientific). For the preparation of a multiplexed microbial SMRTbell library, 1.5 µg bacterial DNA was sheared using g-TUBE (Covaris) at 6000 rpm and purified by AMpure beads PB (Pacific Biosciences). The distribution of sheared DNA was checked by pulsed-field electrophoresis using a pippin pulse electrophoresis system (Sage Science). The multiplexed microbial SMRTbell library was constructed using the "non-size selection" protocol by the manufacturer with SMRTbell Template prep kit 1.0-SPv3 (Pacific Biosciences). Equal amounts of bacterial genome libraries with barcoded adapters were pooled for sequencing. SMRTbell libraries were sequenced on the Pacific Biosciences Sequel platform.

Molecular Plant

Research Article

CellPress

PARTNER JOURNAL

The Reference Genome Sequence of *Scutellaria baicalensis* Provides Insights into the Evolution of Wogonin Biosynthesis

Qing Zhao[1,2,8], Jun Yang[1,2,8], Meng-Ying Cui[1], Jie Liu[1], Yumin Fang[1], Mengxiao Yan[1,2], Wenqing Qiu[3], Huiwen Shang[1], Zhicheng Xu[4], Reheman Yidiresi[4], Jing-Ke Weng[5,6], Tomáš Pluskal[3], Marielle Vigouroux[7], Burkhard Steuernagel[7], Yukun Wei[1], Lei Yang[1], Yonghong Hu[1], Xiao-Ya Chen[1,2] and Cathie Martin[1,2,7]

Genomic DNA was extracted from leaves of a single *S. baicalensis* plant maintained in Shanghai Chenshan Botanical Garden, using a modified CTAB method (Tel-Zur et al., 1999). Quality control was done using a Sage Science Pippin pulse electrophoresis system. Genomic DNA with a length of around 150 kb was sheared using a Megaruptor DNA system and the resulting fragments of 30–50 kb were collected for the

Cell子刊

nature biotechnology ANALYSIS

https://doi.org/10.1038/s41587-019-0266-0

Corrected: Author Correction OPEN

Genomic and phenotypic analyses of six offspring of a genome-edited hornless bull

The integrity of the high-molecular-weight DNA samples was verified on a Pippin Pulse gel electrophoresis system (Sage Sciences). The DNA was then sheared to an average size of 50 kb using a Megaruptor instrument (Diagenode) and verified on a Pippin Pulse gel. A sequencing library was prepared starting

nature plants LETTERS

https://doi.org/10.1038/s41477-018-0289-4

Chromosome-scale assemblies of plant genomes using nanopore long reads and optical maps

resuspended with 3 ml transposable element 10/1 buffer. The extract quality was evaluated using field inverted gel electrophoresis with the Pippin pulse system (Sage Sciences). DNA samples with a fragment size above 50 kb were kept and run on BluePippin (Sage Sciences).

nature communications

Article https://doi.org/10.1038/s41467-022-33530-3

A method for multiplexed full-length single-molecule sequencing of the human mitochondrial genome

2000 (Thermo Fisher Scientific) UV/Vis measurements. To determine the cell line gDNA integrity pulse-field gel electrophoresis, using the Pippin Pulse (Sage Science) was performed. For this analysis a SeaKem® GOLD Agarose 1% (Lonza) gel was prepared in 0.5× TBE buffer

Nature 子刊

SCIENCE ADVANCES | RESEARCH ARTICLE

GENETICS

The Australian dingo is an early offshoot of modern breed dogs

Matt A. Field[1,2], Sonu Yadav[3], Olga Dudchenko[4,5], Meera Esvaran[6], Benjamin D. Rosen[7], Ksenia Skvortsova[2], Richard J. Edwards[3], Jens Keilwagen[8], Blake J. Cochran[9], Bikash Manandhar[9], Sonia Bustamante[10], Jacob Agerbo Rasmussen[11,12], Richard G. Melvin[13], Barry Chernoff[14], Arina Omer[4], Zane Colaric[4], Eva K. F. Chan[2,15], Andre E. Minoche[2], Timothy P. L. Smith[16], M. Thomas P. Gilbert[11,17], Ozren Bogdanovic[2,3], Robert A. Zammit[18], Torsten Thomas[6], Erez L. Aiden[4,5,19,20,21], J. William O. Ballard[22,23]*

(Thermo Fisher Scientific), and molecular integrity was assessed using pulse-field gel electrophoresis. Sage Science Pippin Pulse assessed DNA integrity. A 0.75% KBB gel was run on the 9-hour 10- to 48-kb (80 V) program. DNA ladder used was the Invitrogen 1 Kb Extension DNA Ladder (catalog no. 10511-012). One hundred fifty nanograms of DNA was loaded on the gel.

We generated two libraries that were size-selected on Sage BluePippin gels (Sage Science, Beverly, MA, USA). Libraries were sequenced on Sequel machines with 2.0 chemistry recording 10-hour movies. Sequencing was conducted at the Arizona Genomics Institute, University of Arizona.

Science Advances

PACBIO®

Procedure & Checklist - Using the Sage Science™ Pippin Pulse Electrophoresis Power Supply System

PacBio官方推荐Pippin Pulse

sage science

代表文献

Pippin Pulse + Nanopore

● Long, Meng, et al. "Comparative genomic analysis provides insights into taxonomy and temperature adaption of Aeromonas salmonicida." Journal of Fish Diseases 46.5 (2023): 545-561.

● Keraite, Ieva, et al. "A method for multiplexed full-length single-molecule sequencing of the human mitochondrial genome." Nature Communications 13.1 (2022): 5902.

● Ong, Chian Teng, et al. "Adaptive sampling during sequencing reveals the origins of the bovine reproductive tract microbiome across reproductive stages and sexes." Scientific reports 12.1 (2022): 15075.

● Wunderlich, Stephanie, et al. "Targeted biallelic integration of an inducible Caspase 9 suicide gene in iPSCs for safer therapies." Molecular Therapy-Methods & Clinical Development 26 (2022): 84-94.

● Russo, Alessia, et al. "Low-input high-molecular-weight DNA extraction for long-read sequencing from plants of diverse families." Frontiers in Plant Science 13 (2022): 1494.

● Player, Robert A., et al. "A novel Canis lupus familiaris reference genome improves variant resolution for use in breed-specific GWAS." Life science alliance 4.4 (2021).

● Young, Amy E., et al. "Genomic and phenotypic analyses of six offspring of a genome-edited hornless bull." Nature biotechnology 38.2 (2020): 225-232.

● Belser, Caroline, et al. "Chromosome-scale assemblies of plant genomes using nanopore long reads and optical maps." Nature plants 4.11 (2018): 879-887.

Pippin Pulse + PacBio

● Takeuchi, Tadashi, et al. "Fatty acid overproduction by gut commensal microbiota exacerbates obesity." Cell Metabolism (2023).

● Hadzhiev, Yavor, et al. "The miR-430 locus with extreme promoter density forms a transcription body during the minor wave of zygotic genome activation." Developmental Cell 58.2 (2023): 155-170.

● Kurokochi, Hiroyuki, et al. "Telomere-to-telomere genome assembly of matsutake (Tricholoma matsutake)." DNA Research 30.3 (2023): dsad006.

● Hixson, Kim K., et al. "Annotated genome sequence of a fast-growing diploid clone of red alder (Alnus rubra Bong.)." G3: Genes, Genomes, Genetics (2023): jkad060.

● Field, Matt A., et al. "The Australian dingo is an early offshoot of modern breed dogs." Science Advances (2022): eabm5944.

● Pootakham, Wirulda, et al. "A Chromosome-Scale Genome Assembly of Mitragyna speciosa (Kratom) and the Assessment of Its Genetic Diversity in Thailand." Biology 11.10 (2022): 1492.

● Rahman, M. Julhasur, et al. "LINE-1 retrotransposons facilitate horizontal gene transfer into poxviruses." Elife 11 (2022): e63327.

● Zhao, Qing, et al. "The reference genome sequence of Scutellaria baicalensis provides insights into the evolution of wogonin biosynthesis." Molecular plant 12.7 (2019): 935-950.

Pippin Pulse + Bionano

● Secomandi, Simona, et al. "A chromosome-level reference genome and pangenome for barn swallow population genomics." Cell Reports 42.1 (2023): 111992.

● Dahn, Hollis A., et al. "Benchmarking ultra-high molecular weight DNA preservation methods for long-read and long-range sequencing." GigaScience 11 (2022).

● Palmada-Flores M, Orkin J D, Haase B, et al. A high-quality, long-read genome assembly of the endangered ring-tailed lemur (Lemur catta)[J]. GigaScience, 2022, 11.

● Canaguier A, Guilbaud R, Denis E, et al. Oxford Nanopore and Bionano Genomics technologies evaluation for plant structural variation detection[J]. BMC genomics, 2022, 23(1): 1-17.

● Shukla H, Suryamohan K, Khan A, et al. Near-chromosomal de novo assembly of Bengal tiger genome reveals genetic hallmarks of apex-predation[J]. bioRxiv, 2022.

● Rhie, Arang, et al. "Towards complete and error-free genome assemblies of all vertebrate species." Nature 592.7856 (2021): 737-746.

● Belser, Caroline, et al. “Telomere-to-telomere gapless chromosomes of banana using nanopore sequencing.” Communications biology 4.1 (2021): 1047.

● Zheng, Yuting, et al. "Rapid prenatal diagnosis of Facioscapulohumeral Muscular Dystrophy 1 by combined Bionano optical mapping and karyomapping." Prenatal Diagnosis 40.3 (2020): 317-323.

sage science

参考客户

华大基因 BGI　Novogene 诺禾致源　ANNOROAD 安诺优达　NextOmics 未来组　GrandOmics 希望组

中国科学院昆明动物研究所 KUNMING INSTITUTE OF ZOOLOGY .CAS　复旦大学　中山大学 SUN YAT-SEN UNIVERSITY

中国医学科学院北京协和医学院 Chinese Academy of Medical Sciences & Peking Union Medical College　北京大学　天津大学 TIANJIN UNIVERSITY

军事医学科学院 ACADEMY OF MILITARY MEDICAL SCIENCES　中国农业大学 China Agricultural University　首都医科大学 CAPITAL MEDICAL UNIVERSITY

Sage Science片段回收系列

Pippin Prep™

常规通用型

回收范围：100bp - 1.5kb
样本数量：5个

BluePippin™

全长型

回收范围：100bp - 50kb
样本数量：5个

PippinHT™

高速高通量型

回收范围：100bp - 50kb
样本数量：24个

Pippin 24™

多功能型

片段筛选回收、电泳图谱分析、核酸浓度测定等
样本数量：24个

SageELF™

多通道回收型

回收范围：100bp - 40kb
样本数量：2个

HLS2

超大片段型

回收范围：高达2Mb
样本数量：4个

环亚生物科技有限公司

美国Sage Science大中华区独家代理商

扫码关注微信公众号

地址：上海闵行区友东路358号闵欣大厦1号楼701室
电话：021-54583565
邮箱：info@apgbio.com
网站：www.apgbio.com