# Qsep100<sup>™</sup>全自动核酸蛋白分析系统操作步骤

### 1. Qsep100<sup>™</sup>使用步骤

### 1.1 软件界面

打开 Qsep100<sup>™</sup> 全自动核酸蛋白分析系统主机和<u>空气压缩机</u>的开关,双击软件图标, 点击 New project,设定 Project 名称与存储路径(Browse)(图 1)。

© Q-Analyzer Basic-BETA	
File Edit Tool View Window Setting Language Help	
New project od project Recent project Save project ClogCheck PurgeCheck	
Instrument BIO Control Panel	
Main Method Direct Control	
Project Information	СОМ
User ID New project	Auto 💌
Results Project Directory Project name:	Connect
Sequence Directory	Change Sample
Method Directory	
Comparison Carte	Change Buffer
Cartridge Number	Park
Expiration Date -	
Runs Left	
Real time Last Run Date	
Description	
Sequence Open Save Save as	Run O Stop
SN Sample Method Sample Runs Separation Result Name Para Add	
Position Duration Duration Insert	•••••
1 1 Delete	<b>G</b>
Down	
Down	

图1 新建专案(New project)

### 1.2 连接 Qsep100<sup>™</sup> 与初始化设定

(1) 联机与通气检查

点击 Connect (图 2, A) (仪器图片会从灰色变为彩色,表明联机成功);随之会弹出 Purge Check 提示窗 (图 3-1),<u>点击 Purge check 进行通气检查</u>,根据提示操作:点击 Next (图 3-2) →打开卡夹门→点击 Next (图 3-3) →点击 Purge (30s 后自动停止)(图 3-4) → 点击 Finish (图 3-5)。

ain Method Dire					
meniod Dire	t Control				
	Project Information				СОМ
User ID	zhumimi				Auto
Project Directory	C:\Users\Administrator\Desktop\Result\11	- 9	Latch	(soter, a	A Connect
Sequence Director	C:\Users\Administrator\Desktop\Sequence	-		185-119	Change Sample
Method Directory	C:\Users\Administrator\Desktop\Method		Unlatch		Change Sample
	Cartridge Information	11 -	HV Check		Change Buffer
Cartridae Number			Calibrate	1 martin 1 martin	Park
Expiration Date		1 -		-	
Runs Left		1			
Last Run Date		1			
Description					
Se	quence Open Save	Save as		Run	∩ Stop
N Sample M	ethod Sample Runs Separation Res	sult Name	Add		
N Sample M Position	ethod Sample Runs Separation Res Duration Duration	sult Name	Add		
SN Sample M Position	ethod Sample Runs Separation Res Duration Duration	sult Name	Add Insert	•••••	
SN Sample M Position	ethod Sample Runs Separation Res Duration Duration	sult Name	Add Insert Delete	•••••	H ( ( ( ( ( ( ( ( ( ( ( ( ( ( ( ( ( ( (
SN Sample M Position	ethod Sample Runs Separation Res Duration Duration	sult Name	Add Insert Delete Up		
SN Sample M Position	ethod Sample Runs Separation Res	sult Name	Add Insert Delete Up Down		
Sample M Position	ethod Sample Runs Separation Res	sult Name	Add Insert Delete Up Down 00:00		
N Sample M Position	ethod Sample Runs Separation Res Duration Duration	sult Name	Add Insert Delete Up Down 00:00		
SN Sample M Position	ethod Sample Runs Separation Res Duration Duration	sult Name	Add Insert Delete Up Down 00:00		
SN Sample M Position	ethod Sample Runs Separation Res Duration Duration	sult Name	Add Insert Delete Up Down 00:00	12 11 10 9 8 7 6 Method name:	

图 2 主控页面



图 3-1 联机通气检查



图 3-2





图 3-4

		-	×
1. Please prepare tissues to b NOZZLE" .	lock the " JET-		
2. Then click "PURGE" .(After system will stop purge auto	r 30 seconds the matically).		
		-	
	Stop		
		Previous	Finish

图 3-5

a、点击 Change Buffer (B), P (Park)、W (Wash)、C (Clean) 位置为清洗槽, 放蒸馏水, S 位置放 Separation Buffer (图 4)。

b、在左侧的 MA1 孔中放置 20bp-1000bp 的 Alignment Marker, MA2 孔中放置对应的 Size Marker (C109200-100),在 MB1 孔中放置 20bp-5000bp 的 Alignment Marker, MB2 孔 中放置对应的 Size Marker (C109300-100)(图 5)。用食指和中指托住底部,用拇指将管子 按压到底。

注: P、W、C和S槽中的液面高度以缓冲液槽的刻度线处为宜,不能低于槽体积的 2/3; MA1、MA2、MB1和MB2均取 20-30 µl,再覆盖 20-30 µl 矿物油(Mineral Oil)防止挥发,应避免气泡产生,若有气泡,请离心。

	Project Information			COM
User ID	zhumimi			Auto
Project Directory	C:\Users\Administrator\Desktop\Result\1	1 🗐 🕮 1	Latch	Connect
Sequence Directory	C:\Users\Administrator\Desktop\Sequenc	e -		
Method Directory	C:\Users\Administrator\Desktop\Method		Unlatch	
	Cartridge Information	1	HV Check	B Change Buffer
Cartridge Number			Calibrate	C Park
Expiration Date		. ∏ -		
Runs Left		1		
Last Run Date		· · · ·		
Description				
Se	quence Open Save	Save as		Run O Stop
SN Sample Me	thod Sample Runs Separation	Result Name	Add	
Position	Duration Duration		Insert	•••••
			Delete	•••••
			Up	
			Down	
			00.00	
			00.00	
Micro Vial				12 11 10 9 8 7 6 5 4 3 2 1 SN: Method name:
million viai				SN.   Method hame.

图 4 置换缓冲液和 Marker



图 5 缓冲液和 Marker 位置

(3) 放入样品并复位

点击 Change Sample (A), 放入样品, 关上样品门; 点击 Park (C), 使样品盘复位

### 1.3 卡夹置入与校正(以S2卡夹为例)

(1) 置入卡夹

打开仪器上方的卡夹门,放入卡夹(凹槽向前),关闭卡夹门(图6)。



图 6 置入卡夹

(2) 卡夹锁定和校准

点击 Latch, 卡夹图片颜色变成彩色, 左侧跳出卡夹信息(Cartridge Information), 包括卡夹序号(Cartridge Number)、过期日期(Expiration Date)和剩余可用次数(Runs Left)(图 7)。

**a、若置入的是新卡夹**,则软件会在 Latch 后弹出 Calibration needed 窗口,点击'确定' 进行高压通胶检查(HV check)(图 8-9)。注意:新卡夹需用大头针扎孔后再使用。

Main Method Direc	ct Control Board Setting	
	Project Information	COM
User Type	Advanced	COM9
Project Directory	C:\Users\houze\Desktop\11 Latch	Disconnect
Sequence Directory	y C:\Users\houze\Q-Analyzer\Sequence	Change Sample
moniou birotorij	Cartridge Information	Change Buffer
Cartridge Number	S2-0-160712-2	Park
Expiration Date	2017-JAN-08	
Runs Left Last Run Date Description	200 2016-JUN-12 Standard(Screenin Please finish HV check and calibration before running any seque	ence.
Se	equence Ope	n O Stop
SN Sample M	ethod Sample Runs Separation Result Name Add	

图 7 卡夹锁定



图 8 高压通胶检查信号图

I Chart	
	10.7
	7.0
	7.0
	5.0
	_ ≤
Laboration and Antonion and Antonion	
Manager and the survey of the	2.1
HV Purge tinisned	
HV purge passed, please continue to calibrate cartridge.	-0.8
3倍完 1	
Migration time(min)	
,	

图 9 高压通胶检查通过

高压通胶检查(HV Check)通过后,点击 Calibrate,系统提示是否要进行卡夹校正(图 10),点击'是',系统默认卡夹初次校正的电压为 8KV。卡夹校正成功后点击'确定'(图 11)。

SIO Control Panel							
Main Method Direc	Aain   Method   Direct Control   Board Setting						
	Project Information		СОМ				
User Type	Advanced		сом9 💌				
Project Directory	C:\Users\houze\Desktop\11	Latch	Disconnect	11			
Sequence Directory	C:\Users\houze\Q-Analyzer\Sequence	Liplatch	Change Sample	11			
Method Directory	C:\Users\houze\Q-Analyzer\Method			11			
	Cartridge Information	HV Check	Change Buffer	-111			
Cartridge Number	S2-0-160712-2	Calibrate	Park				
Expiration Date	2017-JAN-08	ł		Ш			
Runs Left	200	Ţ		Ш			
Last Run Date	2016-JUN-12	e		Ш			
Description	Standard(Screening) Do	you want to perform calibration no	ow ?	Ш			
Se	Please make sure you put alignment marker in MA-1 before continuing !           Sequence         Open         S						
SN Sample Me	thod Sample Runs Separatio			1H			
Position	Duration Duration	Insert					
		Delete					

图 10 卡夹校正



图 11 卡夹校正成功

b、若置入的是已用过的卡夹,则在点击 Latch 后,点击 Tool→Recalibrate 进行卡夹重新校正,在弹出的选项框中选择合适的 Voltage(电压)和 Alignment Marker(内参)类型, 点击 Start Calibration(图 12),校正完成时会弹出 Calibration succeed 窗口,点击确定(图 13)。



图 12 卡夹重校正(Recalibration)



图 13 卡夹重校正成功

### 1.4 程序设定(图 14)(以 S2 卡夹为例)

(1) 单击 Sample Position 下方的空白格(A),弹出 96 孔盘模拟图(图 15),于左侧 选择所放样品位置,可单选,也可点击右侧 A-H 或下方 01-12 坐标以直接选取一行或一列, 双击右侧相应位置处输入样品信息(Sample ID),设置完成后点击 OK;

(3)单击 Method 下方的空白格 (B),弹出测试方法选择框(图 16),选择适合的 Alignment Marker 和 Method (注意: 所选方法的电泳电压要与卡夹校正时的电压一致), 选好后点击 OK; Sample Duration (吸样时间)、Runs (检测重复次数)和 Separation Duration (分离时间)可以根据样本情况灵活设定 (双击输入);

(4) 在 Result Name 里输入一批样品的名字,在下面选项框中选择 Sample ID;

(5) 点击小计算器图标(C), 弹出 Calculate Flow 框, 文库样本的建议参数如图 17 所示。勾选 Calculate, 点中 Create size marker, 下面的 Size marker injection time 选择 5s, 点击 OK (为保证实验结果的准确性,建议每天或每批样本新建一个 Size Marker);

(6)程序设好后,点击 Run,开始电泳分离(图 18)。使用者可点击 Real time 实时监测电泳过程。 注意:点击 Run 后不能打开样品门,否则运行自动停止!

File Edit To	ool Vie	w Window	v Setting La	anguage	Help							
	New p	roject Load	project Recen	P It project S	ave proj	ect Recal	Jibrate C	ClogCheck	PurgeC	heck		
<u></u>	BIO	Control Pan	el									
Instrument	Ma	in Method	Direct Contro	i)								
			Pr	oject Inf	ormati	on						СОМ
	U	ser ID			zhumin	ni						Auto 💌
Results	P	roject Direc	tory		D:\\Res	ult\161018			10	Latch	CISTER REAL	Connect
AL	S	equence Dir ethod Direc	rectory tory		D:\\Seq	uence				HV Check		Change Sample
			Car	tridae Ir	format	tion				Calibrate		Change Buffer
Comparison												Park
RealTime		artridge Nur	nber						TT.			
	E	kpiration Da	te						Ý			
	R	uns Left							1		Contraction of the second seco	
Real time	Li	ast Run Date	e									
	D	escription										
			Sequen	ce	Open	Sa	ve	Save a	s		D Run	Stop
	St	Sample	Method	Sample	Runs	Separation	Res	ult Name	Para	Add		
		Position		Duration		Duration				Insert		ООООН
	1				1		None		- E	Delete		
		- <b>A</b> -	B				Inone			Lin		
										Up		
										Down		

图 14 程序设定



图 15 样品位置选择

Method Selector			83
Application (	ORA CRNA	C Glycan	C Protein
Alignment Marker	✓ MA-1 ▼ 20 1	000 C Reduce • M	Normal C Enhance
Cartridge Type	S2 💌 Standard cartrid	lge(Shelf Life: 6 Months)	
Sample concentration (	C High (> 10 ng/ul) (4	Regular (0.1 ~ 10 ng/ul) 🤇	C Low (< 0.1 ng/ul)
Method	Description	Range	Remark
M-4-10- <mark>08</mark> 200	Sample injection 4kv 10s Separation 8kv 200s	15~5000 bp Best resolution: 4~10 bp	
M-4-10-10-150	Sample injection 4kv 10s Separation 10kv 150s	15~5000 bp Best resolution: 10~50 bp	
gDNA	Sample injection 4kv 10s		Up to 15Kbp genomic DNA
gDNA(NGS)	Sample injection 4kv 10s		Sheared genomic DNA
T-HvPurge-08-120	Gel refill with HV on for		
T-Purge-120	Gel refill without HV for		
High voltage purge (	C Purge 🔲 Pu	rge Modification	
Customized Method			ок

图 16 电泳方法选择

<ul> <li>Calculate Flow</li> </ul>	v	
Baseline Fac     Peak Thresho	tor: 200	
Calculate	C Default	Browse
	Size marker injection time: 5 sec(s)	
	User defined reference maker table:	
		Browse
C Smear	C Distribution 100 + % C Range ~ bp	
Peak Calling		Browse
🗖 Diagnosis		Browse
🗖 Auto Recalibr	ate	
🗖 Auto Assign 1	18S 28S	
		OK Cancel

图 17 Size marker 的选择

Control Panel		
Main Method Direct Control Board Setting		
Project Information		СОМ
User Type Advanced		COM9 💌
Project Directory C:\Users\houze\Desktop\11	Latch	Disconnect
Sequence Directory C:\Users\houze\Q-Analyzer\Sequence		
Method Directory C:\Users\houze\Q-Analyzer\Method	Unlatch	
Cartridge Information	HV Check	Change Buffer
Cartridae Number 82.0.160710.0	Calibrate	Park
Calchage Number 52-0-100/12-2		
Runs Left 200		
Last Run Date 2016-AUG-28		
Description Standard(Screening)		
Sequence Open Save Save as		C Run O Stop
SN Sample Method Sample Runs Separation Result Name	Add	
Position Duration Duration	Insert	••••
1 A-01,A-0 M-8-10-08-200 10 1 200 文库样本 Sample ID 🗾	Delete	
	Up	
	Down	
	00.28	••••••••••••
	00120	•••••
Micro Vial		SN: Method name:
Reduce 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1 1		Extend 0
-100 -80 -00 -40 -20 0	20 4	•0 00 80 100

图 18 启动程序

## 2 数据结果分析

## 2.1 打开检测结果

样品检测完成后,点击 Open file,选中目标结果文件,打开(图 19-20)。

File Edit T	Fool View Window Setting	Language Help			
	Open file Recent file Save file	Save all Calculate Smear Peak Calling Para	ameters Show BP/L	egend Show BP/N	lin Invert
Instrument	Filename	Þ			
	<b>0</b> Open File				23
		25-吉因加 -	• ◆• 搜索 20160	725-吉因加	P
	组织 ▼ 新建文件夹				0
Results		名称 ^	修改日期	类型	•
FALL		10s_1_S1B02_R1	2016/7/25 17:22	BOPX 文件	
		🖻 10s-repeat_1_S1B02_R01	2016/7/25 17:22	BOPX 文件	
	·····································	10s-repeat_1_S1B02_R02	2016/7/25 17:22	BOPX 文件	
Comparison		10s-repeat_1_S1B02_R03	2016/7/25 17:22	BOPX 文件	
RealTime		10s-repeat_1_S1B02_R04	2016/7/25 17:22	BOPX 文件	- 11
A n n		10s-repeat_1_S1B02_R05	2016/7/25 17:22	BOPX 文件	=
	▶ 🛗 视频	10s-repeat_1_S1B02_R06	2016/7/25 17:15	BOPX 文件	
Real time	▷ 🔛 图片	10s-repeat_1_S1B02_R07	2016/7/25 17:22	BOPX 文件	
i tour unito	▶ 📑 文档	10s-repeat_1_S1B02_R08	2016/7/25 17:26	BOPX 文件	
	> 👌 音乐	10s-repeat_1_S1B02_R09	2016/7/25 17:32	BOPX 文件	
		10s-repeat2_1_S1B02_R1	2016/7/25 17:39	BOPX 文件	
	▲ 1 및 计算机	SizeMarker 5s MA2 S1MA2 R1	2016/7/25 16:28	BOPX 文件	-
	~ *				•
	文件	名(N): "10s-repeat2_1_S1B02_R1" "10s-repeat_	_1 ▼ Result file 20	)14(*.bopx)	-
			打开(O)	取消	

图 19 打开结果文件



图 20 结果文件显示框

### 2.2 新建比对档

点击 Comparison→New Folder,用鼠标将计划处理的结果拖进 New folder 框内(图 21)。 勾选 Time Alignment (时间对齐),点击 Apply all;勾选 Signal Alignment (信号基底值对 齐),点击 Apply all; Signal offset 调为'-1',点击 Apply all (可点击多次),调整结果间距 (图 22)。



图 21 新建比对档 (New Folder)



<b>o</b> Fo	lder 1										
Sign	al Gel View										
	B-02	B-02	B-02	B-02	B-02	B-02	MA-2				Offent
-										Os-repeat 1 S1B	Oliset
									2 1	0s-repeat_1_S1B	23535
									3 1	0s-repeat_1_S1B	47125
									4 1	0s-repeat_1_S1B	70721
									5	0s-repeat_1_S1B	94343
									6 1	l0s-repeat_1_S1B	117889
										Sizemarker_5s_MA	1415/1
								1			
									Chart a	ittributes	
									🔽 Sig	gnal Alignment 🛛	Apply All
									. <u>™</u> 11	ne Aignment	Apply All
									Global	attributes	
									Signal		
								-	orgital		
										R	eset All

图 22 结果比对(峰图&胶图)

#### 2.3 导出数据

(1) 导出峰图和胶图

于空白处右击,选择 Export signal chart,导出峰图。点击左上角的 Gel View,可展示 模拟胶图,于空白处右击,选择 Export gel view,导出胶图(图 23)。

(2) 导出报告

于空白处右击,选择 Export report,选择导出报告的格式(Report Type);若检测的是 PCR 产物,则选择 Standard(DNA),若检测的是文库样本,需要 Smear 信息,则选择 Smear, 若检测的样品为 RNA,则选择 RNA,若有建立 Peak calling table 信息,则选择 Peak calling。 再保存即可(图 24)。



#### 图 23 导出结果图片



图 24 导出报告

## 附: 5K Size marker 的构建(以 S1 卡夹为例)

(1) 在 Latch 卡夹后,进行卡夹校准时, Alignment marker 需选择 MB-1 (如下图 1)。

Recalibration		1.1		×
C 10KV	Marker Position (© MA-1(20 - 1K) (© MB-1(20 - 5K)	Alignment Marker Last Calibrated Date 2016-AUG-28 N/A	LM_SN 371.63 N/A	UM_SN 366.63 N/A
	Start Calibrati	on Cancel		

图 1 5K marker 建立时的卡夹校准

(2) 校准完成后,Add 一个新程序(图 2),点击 Sample Position 下方空白格子(B处), 在弹出的窗口中选中 MB2 孔(图 3),点击 OK。



图 2 添加新程序



图 3 MB2 孔的选择

(3)点击 Method 下方空白格子(C处),弹出检测方法选择框(图4),Alignment Marker 选择 MB1,选择 Method 时也同样注意电压要与卡夹校准时的电压一致,选好后点击 OK; Sample Duration(吸样时间)设置为 5s、其他默认;就可以点击 Run。

Method Selector	- 1/5	e/11.	×			
Application (		A C Glycan	C Protein			
Alignment Marker 🛛	MB-1 💌 20	5000 C Reduce	Normal O Enhance			
Cartridge Type S	1 💌 High resoluti	on cartridge(Shelf Life: 6 I	Months)			
Method	Description	Range	Remark			
M-4-10-06-300	Sample injection 4kv	15~1000 bp Best resolution: 2~4 bp	<u>_</u>			
M-4-10-06-500	Sample injection 4kv	15~15k bp Best resolution: 2~4 bp				
M-4-10-08-240	Sample injection 4kv	15~5000 bp Best resolution: 4~10				
M-4-10-10-150	Sample injection 4kv	15~5000 bp Best resolution: 10~50				
M-8-10-06-300	Sample injection 8kv	15~1000 bp Best resolution: 2~4 bp	For low concentration			
M-8-10-06-500	Sample injection 8kv	15~15k bp Best resolution: 2~4 bp	For low concentration			
M-8-10-08-240	Sample injection 8kv	15~5000 bp Best resolution: 4~10	For low concentration			
High Voltage Purg	je C Purge	Purge Modification				
Customized Metho	Customized Method OK					

(3) 结果处理

以上程序完成后得到的结果图(图5),只有峰型,没有片段 bp 大小,需手动赋值。 a、先删除 A 区数据栏里的杂峰(RFU 值小于 4 的均是杂峰),也可通过挨个点击,看箭头 所指来确定是否为杂峰。



图 5 MB2 初始结果

b、点击 Edit→Ref Marker (图 6), 弹出图 7, 点击左上角 Load, 弹出软件自带的 所有 Marker 组合,根据所用的卡夹类型,所用电压和选用的 Marker 选择对应的 Ref Marker (图 8), 此处以 S1-6-S109300-20-5K. ref 为例:

(表示 S1 卡夹, 6KV 电压下 5K 的 Size Marker 和 5K 的 Alignment Marker 组合下跑出的结果);



3 <b>0</b> Q-	• Q-Analyzer Basic								
File	Edit	Tool	View	Window	Setting	Language	Help		
	Sample file								
RefMarker									
	Pa	aramet	ers						
	Peak Calling Table		ble						

图 6 点击 Ref Marker

BIO Refe	erence Ma					
Loa	d	Save as				
Conce	ntration c	orrection factor 1.00	Length Factor 1.00			
No	No Time Peak Area			Concentration(ng/µl)		

图 7 Reference marker editor



图 8 选择合适的 Reference Marker 文件打开

c、选中对应的 Ref Marker 打开后得图 9, Ref Marker 为 17 条带,需与图 5 (A 区) 的条带数一致,于图 5 (A 区) 任意处右击,选择 Copy reference marker data,到图 9 数据栏任意处右击,选择 Paste reference marker data,即可将新跑的 MB2 的出峰时间 和信号值粘贴到具有片段大小的 Ref Marker 里,构建出当下实验可用的 MB2 (图 10),再 右击图 10 左上方的 Save as,将建好的 MB2 数据另存为新的 5k 的 Size Marker,用于后 面样品产物大小的计算。

Bio	BIO Reference Marker Editor - S1-6-C109300-20-5K.rfm 📃 📃 🔽								
	Load	Save as							
Co	Concentration correction factor 1.00 Length Factor 1.00								
No	Time	Peak Area	bp	Concentration(ng/µl)					
1	113.96	63853	20.00	0.53					
2	126.08	57206	50.00	1.00					
3	141.36	22391	100.00	0.50					
4	156.84	25837	150.00	0.50					
5	172.40	27661	200.00	0.50					
6	187.80	24455	250.00	0.50					
7	201.24	52999	300.00	0.85					
8	213.32	31291	350.00	0.50					
9	221.12	22070	400.00	0.40					
10	225.24	14169	430.00	0.25					
11	228.28	23625	450.00	0.40					
12	233.56	28361	500.00	0.50					
13	245.12	64521	750.00	0.60					
14	255.16	68123	1100.00	0.80					
15	263.36	100752	1800.00	1.20					
16	272.60	107744	3000.00	0.75					
17	281.24	165258	5000.00	0.56					

图 9 选中的 Reference Marker 文件的具体信息

BIO	Reference Marker Editor - S1-6-C109300-20-5K.rfm 📃 🗖 🔽							
	Load	Save as						
Со	Concentration correction factor 1.00 Length Factor 1.00							
No	Time	Peak Area	bp	Concentration(ng/µl)				
1	117.28	542648	20.00	0.53				
2	126.04	486439	50.00	1.00				
3	139.28	296564	100.00	0.50				
4	153.36	337628	150.00	0.50				
5	168.08	395325	200.00	0.50				
6	182.56	332862	250.00	0.50				
7	195.76	689470	300.00	0.85				
8	206.20	402268	350.00	0.50				
9	213.68	318287	400.00	0.40				
10	216.84	172455	430.00	0.25				
11	220.48	325073	450.00	0.40				
12	225.32	385911	500.00	0.50				
13	236.28	760173	750.00	0.60				
14	244.88	1045637	1100.00	0.80				
15	252.40	1599654	1800.00	1.20				
16	261.20	1441585	3000.00	0.75				
17	271.96	2178001	5000.00	0.56				

图 10 新构建的 MB2 的结果