



Recycle Bin



Data



LibreOffice
6.2



MO.Control 2
(x86)



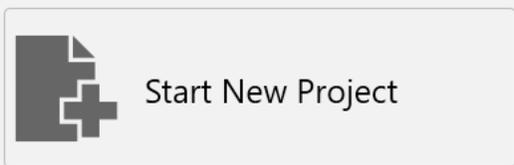
TeamViewer



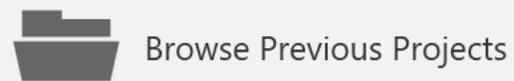
SunloginClie
nt

← 1. 打开MO. Control 2 控制软件并打开仪器右后方的开关

NOTEMPER



← 2. 点击New Project



Recent:

VCB_MZ1 - 2023/11/27 16:57:20

C:\Users\NTC\Desktop\DATA\训练moc2

20221019DA019-hepes -

C:\Users\NTC\Desktop\yituo

20221019DA019 -

C:\Users\NTC\Desktop\yituo

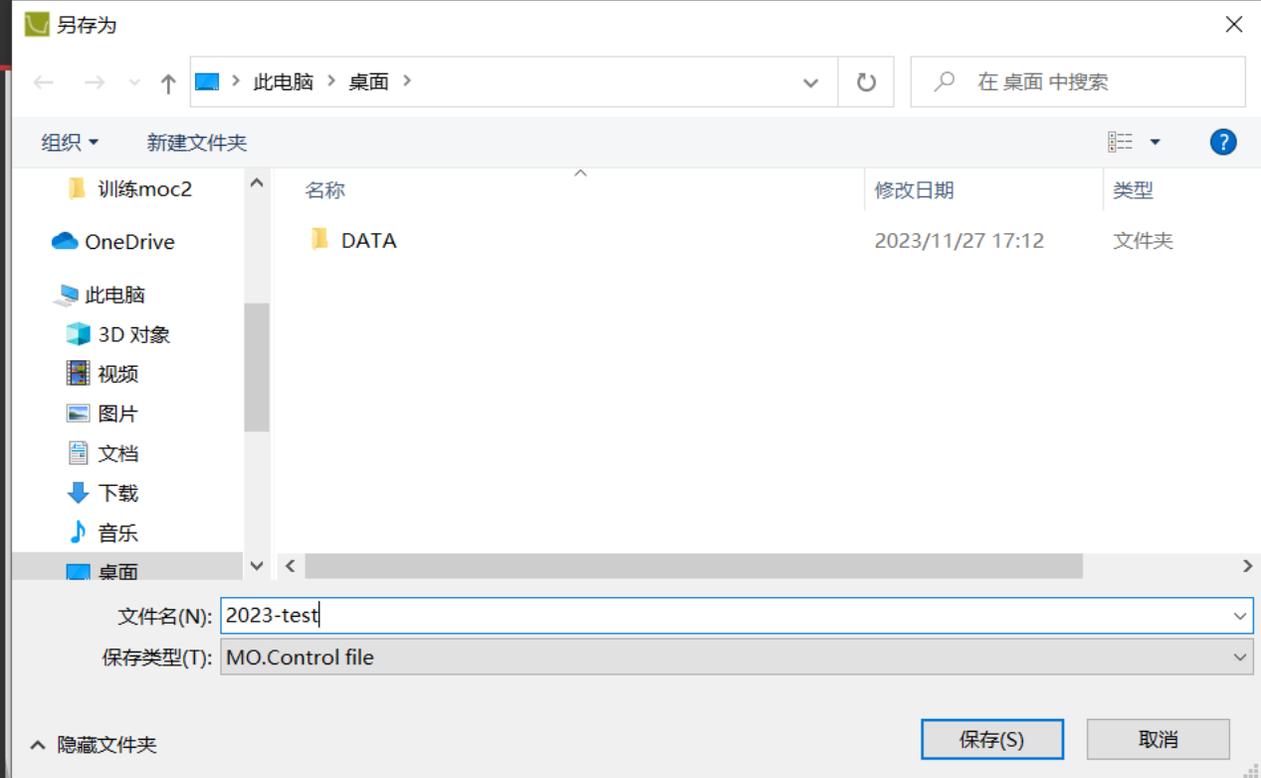
20221019 -

C:\Users\NTC\Desktop\yituo

20221018 -

C:\Users\NTC\Desktop\yituo





New Project

Previous Projects

3. 填写project 名称，点保存，project建立即实验moc2原始文件建立

- Recent:
- VCB_MZ1 - 2023/11/27 16:57:20
C:\Users\NTC\Desktop\DATA\训练moc2
 - 20221019DA019-hepes -
C:\Users\NTC\Desktop\yituo
 - 20221019DA019 -
C:\Users\NTC\Desktop\yituo
 - 20221019 -
C:\Users\NTC\Desktop\yituo
 - 20221018 -
C:\Users\NTC\Desktop\yituo



Project Overview

Monolith X

Change

Ratio 670nm / 650nm

Change

Guidance



Close



Save

Please select a binding mode

Get online help

Binary binding



Ternary binding



Protein Labeling Assistant



Expert Mode



4. 点击Binary binding, 进入互作实验测定。



Experiments (0)

Merge sets (0)

Comparisons (0)

New

Project
Overview

Monolith X

Change

Ratio 670nm / 650nm

Change

Binary binding

Change

Guidance



Close



Save

Please select an experiment

Get online help

Pretest



Binding Check



Binding Affinity

Protein Labeling Assistant

Expert Mode



5. 点击Binary Affinity，直接进行亲和力测定实验。

^ Experiments (0)

Merge sets (0)

Comparisons (0)

+ New



搜索



14°C 局部多云

17:54
2023/11/27

Project Overview

E1 Binding Affinity

1 Plan 2 Instructions 3 Data 4 Details

Stable at 25°C Change Ready

Close Save

Plan Your Experiment

You can enter here some comments about this experiment

实验备注

Target

MyTarget

记录Target名称

Assay buffer

MyBuffer

记录所用的交互Buffer名称

Experiments (1)

E1 Binding Affinity

Experiment 1

记录实验名称

Use His-Tag Labeling

Concentration of stock solution

记录Target的储液浓度及互动时实验浓度

20 nM

Capillary

Monolith Premium Capillary

记录所用的毛细管

Concentration in this assay

Ligand

MyLigand

记录Ligand名称

Estimated Kd

optional

Concentration of stock solution

记录Ligand的储液浓度

μM

Ligand in organic solvent like DMSO

Ligand buffer in this assay

Highest concentration in this assay

记录Ligand互动时实验浓度(点后面小笔可以填写)

-

Excitation

IR Laser Power

Auto-detect

Medium

6. 填写数据信息，进行实验记录。

Go to Instructions



Project Overview

Close Save

E1

Binding Affinity

1 Plan 2 Instructions 3 Data 4 Details

Aptamer 20 nM	AMP 5 mM	C Buffer Capillary	Auto-detect excitation Medium Power	Temperature	<i>You can enter here some comments about this experiment</i>
------------------	-------------	-----------------------	--	-------------	---

Plan Your Experiment

Target

Aptamer [dropdown] [?]

Use His-Tag Labeling [?]

Concentration of stock solution 40 nM [?]

Concentration in this assay 20 nM [edit] [?]

Ligand

AMP [dropdown] [?]

Estimated Kd optional μM [?]

Concentration of stock solution 10 mM [?]

Ligand in organic solvent like DMSO [?]

Ligand buffer in this assay 50.0% [?]

Highest concentration in this assay 5 mM [edit] [?]

Assay buffer

C Buffer [edit] [?]

Capillary Monolith Capillary [dropdown] [?]

Excitation

Auto-detect [edit] [?] Medium [edit] [?]

IR Laser Power

Experiments (1)

E1 Binding Affinity

Aptamer-AMP [edit]

Merge sets (0)

Comparisons (0)

+ New [dropdown]

7. 填写完成后, 点击Go to Instructions, 进入仪器指导界面

Go to Instructions

Project Overview

Close Save

Experiments (1)

E1 Binding Affinity

Aptamer-AMP

Merge sets (0)

Comparisons (0)

+ New

E1 Binding Affinity

1 Plan 2 Instructions 3 Data 4 Details

Stable at 25°C Change Ready

Aptamer 20 nM AMP 5 mM C Buffer Auto-detect excitation Temperature

Capillary Medium Power

You can enter here some comments about this experiment

仪器指导界面是仪器根据您填写的信息提供的样品稀释方案及混合互动指导。若您样品已稀释准备好,则可直接进行倍比稀释,混合互动后毛细管取样。

Instructions

Below are detailed instructions on how to prepare the samples necessary for your experiment. For general information on binding affinity experiments, refer to the guidance on the right.

The minimum required stock solution and buffer volumes for this experiment are:

- 200 µl target stock solution (40 nM of Aptamer)
- 30 µl ligand stock solution (10 mM of AMP)
- 200 µl buffer of the ligand stock solution (ligand buffer)

1. Start by preparing the following intermediate samples:

- a. No predilution required. Use 200 µl of 40 nM Aptamer stock solution for the following steps.
- b. No predilution required. Use 30 µl of AMP for the following steps.
- c. No predilution required. Use 200 µl of ligand buffer for the following steps.

2. Prepare a serial dilution of the ligand using the ligand buffer.

Remember to discard 10 µl of the lowest concentration sample in tube 16 to get an equal volume of 10 µl in all samples.

More info

3. Add 10 µl of 40 nM Aptamer to each tube from 16 to 1 and mix by pipetting.

More info

4. Dip a Monolith capillary into each tube from 1 to 16, put them in

Open drawer

Back

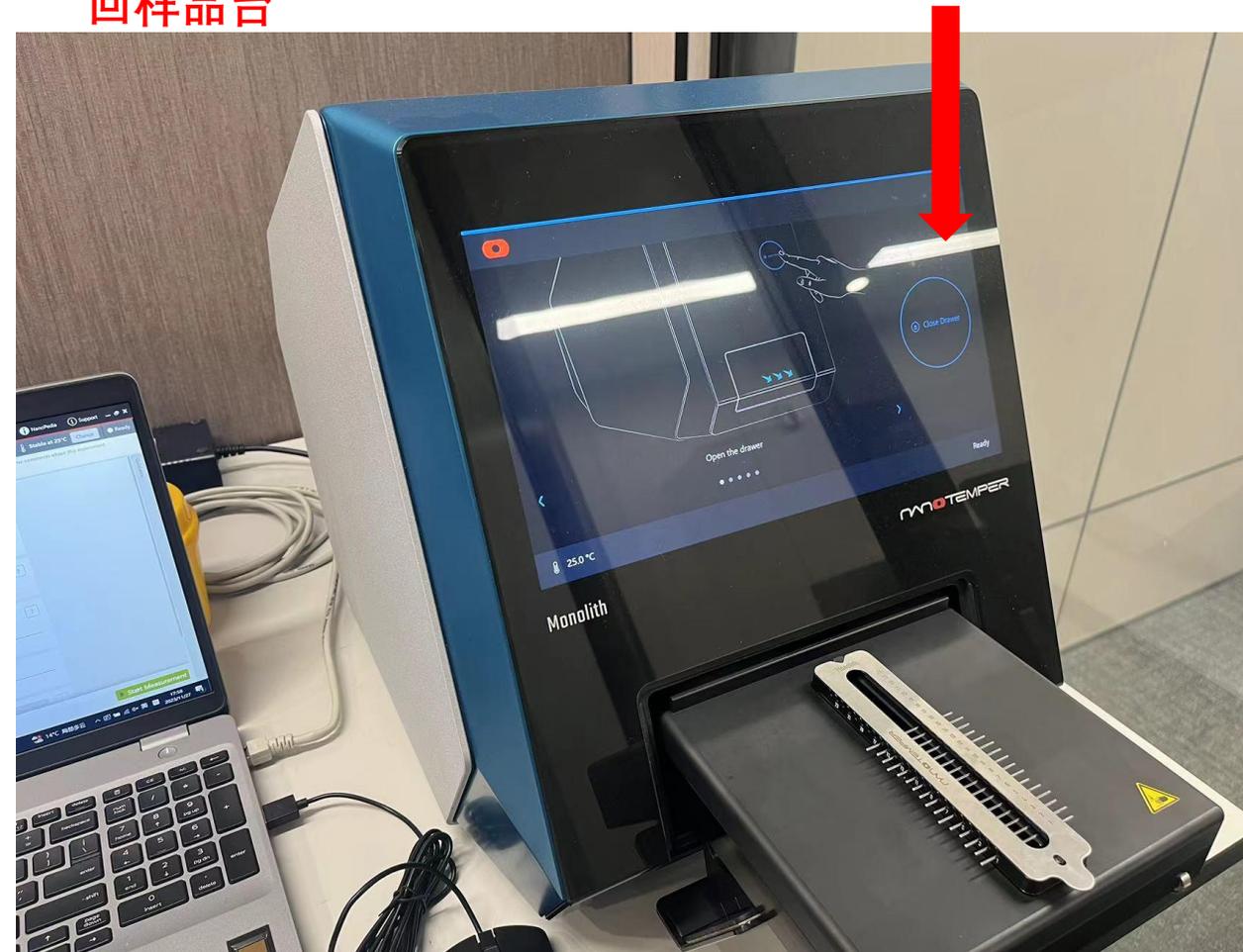
Print Instructions

Start Measurement

8. 触屏点击Open Drawer，打开样品台



9. 放置好毛细管后，触屏点击Close Drawer，仪器收回样品台



Project
Overview

E1 Binding Affinity

1 Plan

2 Instructions

3 Data

4 Details

Stable at 25°C

Change

● Ready

Aptamer
20nMAMP
5mM

C Buffer

Auto-detect excitation

Temperature

You can enter here some comments about this experiment

Capillary

Medium Power



Close



Save

Experiments (1)

E1 Binding Affinity

Aptamer-AMP

Merge sets (0)

Comparisons (0)

+ New

Instructions

Below are detailed instructions on how to prepare the samples necessary for your experiment. For general information on binding affinity experiments, refer to the guidance on the right.

The minimum required stock solution and buffer volumes for this experiment are:

- 200 μ l target stock solution (40 nM of Aptamer)
- 30 μ l ligand stock solution (10 mM of AMP)
- 200 μ l buffer of the ligand stock solution (ligand buffer)

1. Start by preparing the following intermediate samples:

- No predilution required. Use 200 μ l of 40 nM Aptamer stock solution for the following steps.
- No predilution required. Use 30 μ l of AMP for the following steps.
- No predilution required. Use 200 μ l of ligand buffer for the following steps.

2. Prepare a serial dilution of the ligand using the ligand buffer.

Remember to discard 10 μ l of the lowest concentration sample in tube 16 to get an equal volume of 10 μ l in all samples.



More info

3. Add 10 μ l of 40 nM Aptamer to each tube from 16 to 1 and mix by pipetting.



More info

4. Dip a Monolith capillary into each tube from 1 to 16, put them in

Open drawer

Back

10. 样品放置完成后, 点击Start Measurement, 仪器开始运行, 进行检测

Print Instructions

Start Measurement



搜索



14°C 局部多云

17:59
2023/11/27

Project Overview

E1 Binding Affinity

1 Plan 2 Instructions 3 Data 4 Details

Stable at 25°C Change

Working 11/27/2023

Aptamer 20nM

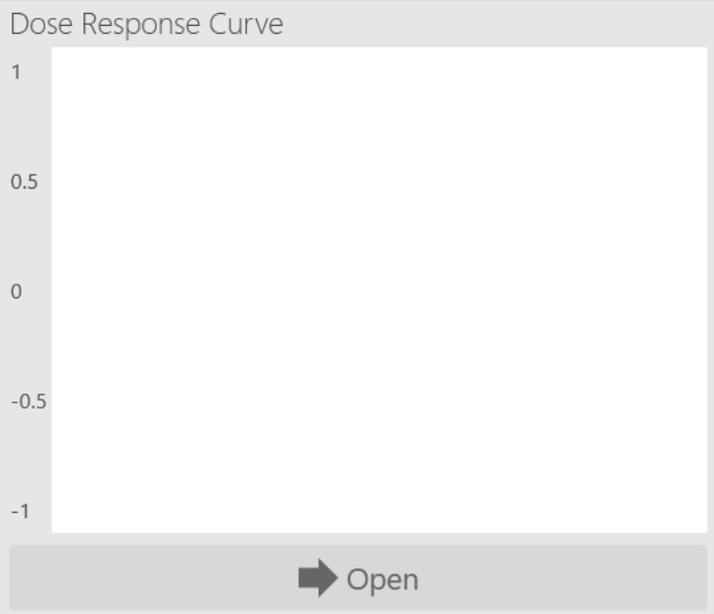
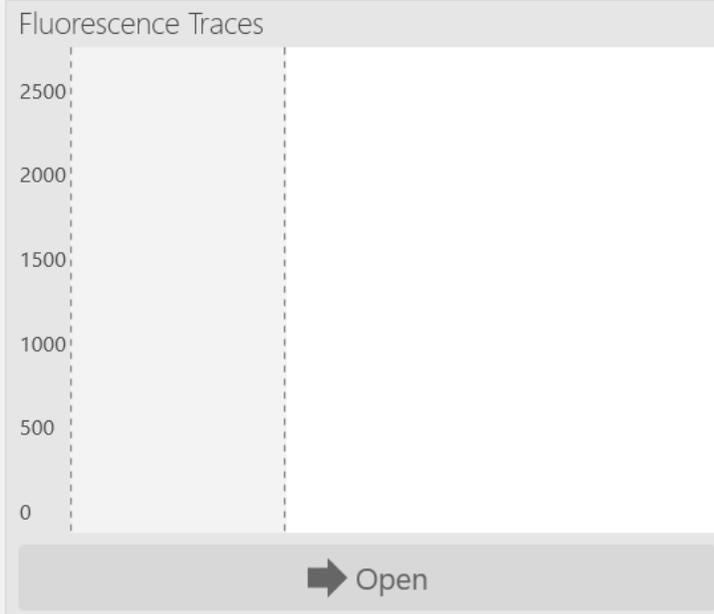
AMP 5mM

C Buffer Capillary

unknown, auto-detect... Medium Power

Temperature 25.0°C

You can enter here some comments about this experiment



Progress

Live View

0%

Auto-detecting excitation power approximately 06:37 remaining

11. 整个亲和力测定为6分半左右，会实时显示测定的数据情况

Back

Cancel measurement

Project
Overview

E1 Binding Affinity

1 Plan 2 Instructions 3 Data 4 Details

Stable at 25°C Change Completed 11/27/2023

Aptamer
20nMAMP
5mM

C Buffer

50% excitation

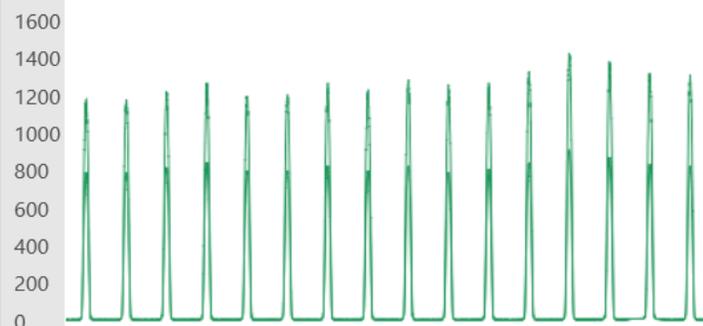
Temperature
25.0°C

Capillary

Medium Power

You can enter here some comments about this experiment

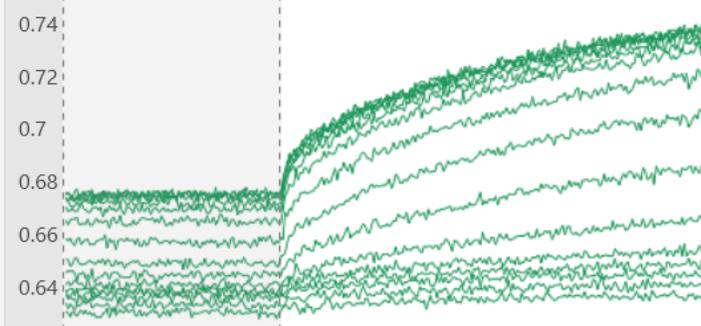
Capillary Scans



- Fluorescence intensity
- No fluorescence variation
- No adsorption

Review

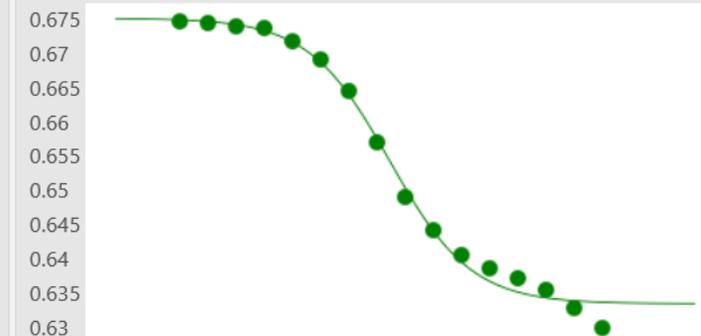
Fluorescence Traces



- No aggregation
- No photobleaching rate changes

Review

Dose Response Curve



- Signal/Noise ratio is large enough to detect binding

Review

Result

 $K_d = 26.9 \mu\text{M}$

Things to check

If there are not enough data points at either the low ligand concentration or the high ligand concentration ends, consider adjusting the starting concentration of your ligand

12. 测定完成后，点击+Binding Affinity，软件会加载之前填写的参数，便于进行重复实验，或者+New 选择 Experiment开始新的实验。

Back

+ Binding Affinity

+ New