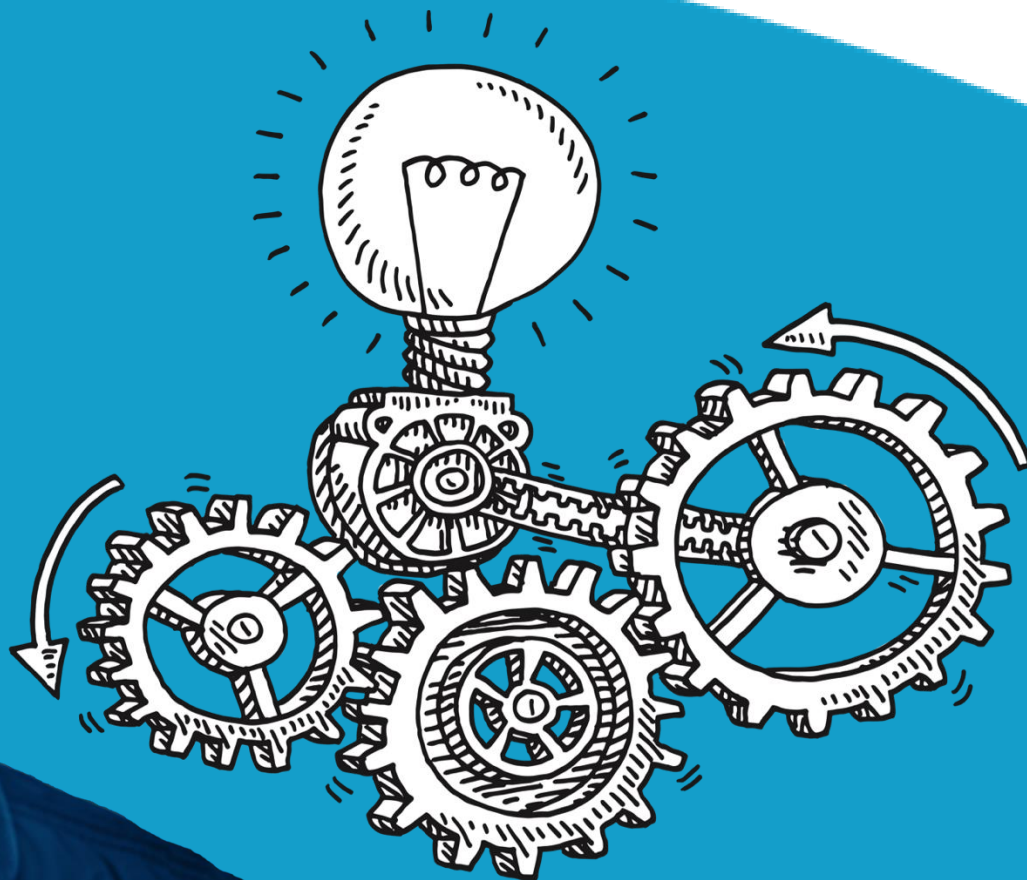


# Simple Plex 技术原理概述



# Epic Tools for Epic Science

## biotechne®



R&D SYSTEMS

ACD

NOVUS  
BIOLOGICALS

TOCRIS

R&D SYSTEMS  
CLINICAL CONTROLS

RNA  
MEDICAL

CLINIQA

biospecific

BIONOSTICS  
Quality Solutions

蛋白质分析仪器

生命科学

临床诊断

# Epic Tools for Epic Science

protein simple<sup>®</sup>  
a biotechne brand

## 纯化的蛋白质 (生物制药)

### ICE

*iCE-3*



全柱成像  
cIEF  
光吸收检测

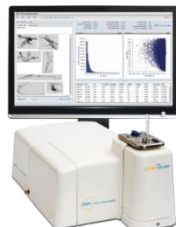
*Maurice*



- 全自动
- 全柱成像 cIEF
- CE-SDS
- 光吸收和自发荧光检测

### MFI

*5100/5200*



- 蛋白团聚体
- 颗粒成像

## 非纯化蛋白质(裂解物等)

### Imaging



紫外-可见-  
红外凝胶成  
像和化学发  
光成像

### Simple Western

*Wes and the Sues*



- 全自动分子量和/或电荷分析
- 蛋白捕获
- 免疫检测

### Simple Plex

*Ella*

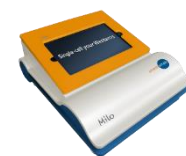


- 全自动
- 多因子分析
- 超灵敏
- 微流控 ELISA

## 细胞

### Single Cell Western

*Milo*



- 单细胞级别 Western
- 种群分析

# Ella

- 体积小巧

- 尺寸小 – 37 cm (w) x 54 cm (d) x 26 cm (h)
- 重量轻 – 16 kg
- 无废液（仪器内部）

- 简单易用

- 即插即用
- 经过验证的 cartridge
- **15** 分钟加样，**1** 小时出结果



# 全自动微流控免疫检测设计

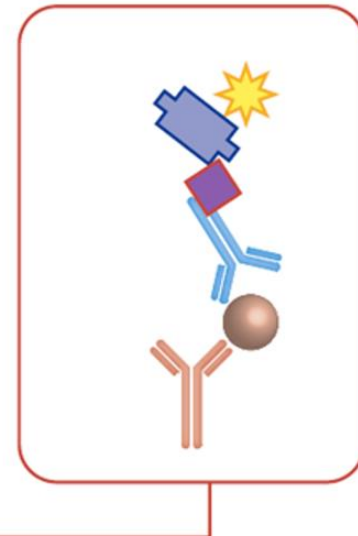
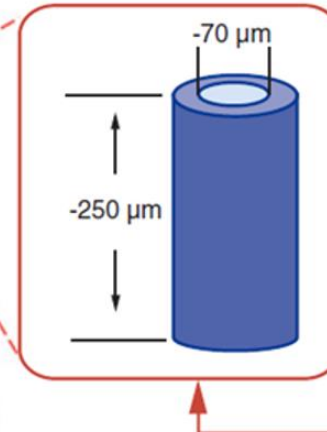
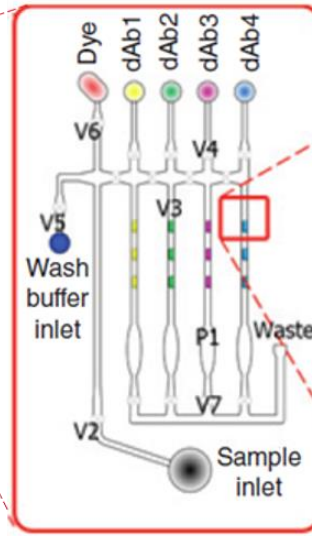
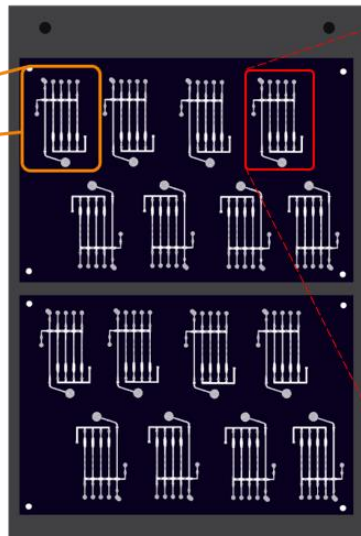
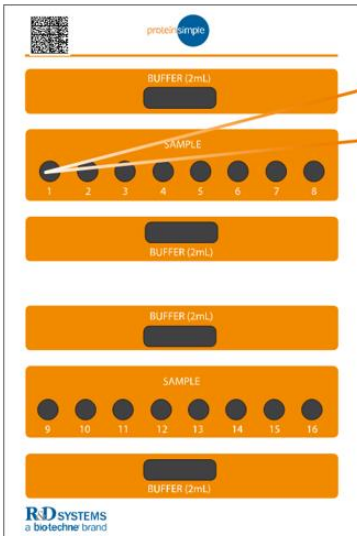
Simple Plex Cartridge

正面

背面

微流路

包被有抗体的GNR



1. 原理：双抗夹心
2. 抗体对来自R&D system，高亲和力，高特异性
3. 全封闭检测空间，降低噪音
4. Biotin-Streptavidin信号放大系统
5. 荧光法高线性范围
6. 微流控：节省珍贵样本
7. 全自动检测：实现标准化

# 检测只需三步



## 扫描毛细管板二维码

- 标准曲线
- 毛细管靶蛋白包被信息
- 生产日期，批号等



## 样本准备

- 5 分钟
- 25  $\mu$ L sample + 25  $\mu$ L diluent
- 1 mL x 9 assay buffer (72x1)
- 2 mL x 4 assay buffer (16x4)



## Place cartridge in ELLA

- 输入样品信息
- 开始实验
- 一小时完成检测

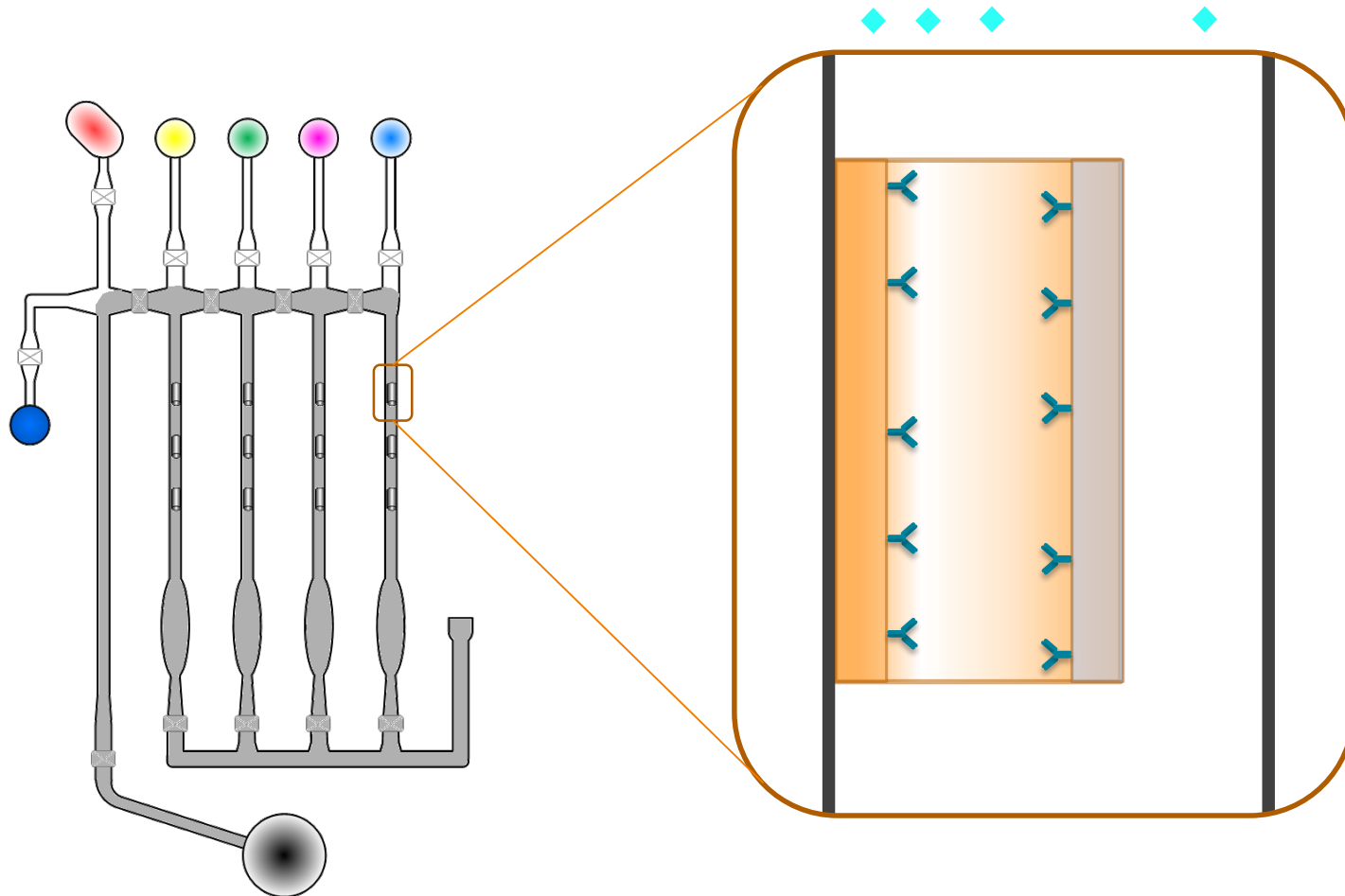
5分钟手工操作，自动化检测  
实现检测标准化

# How Simple Plex works

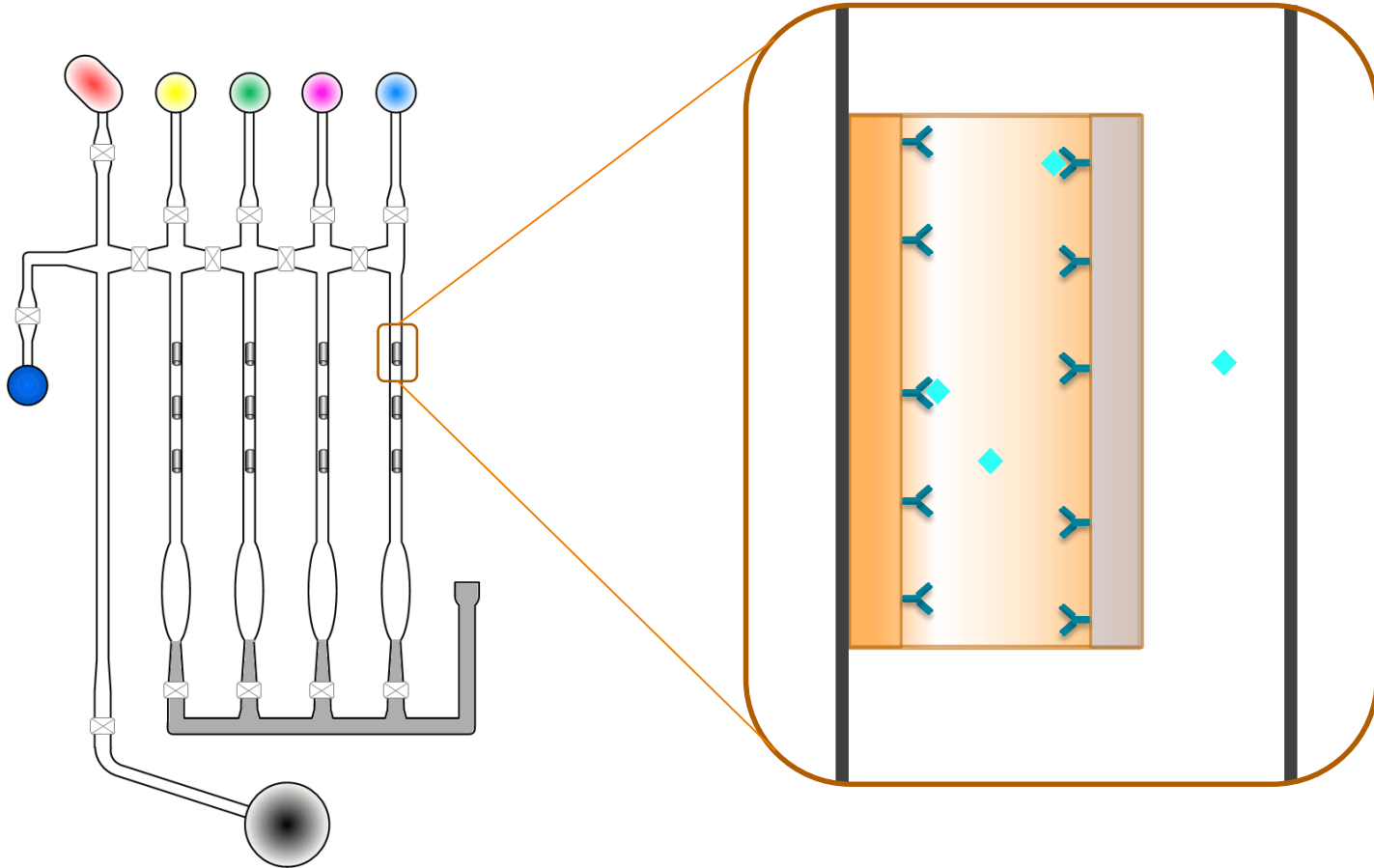


Ready to Ship

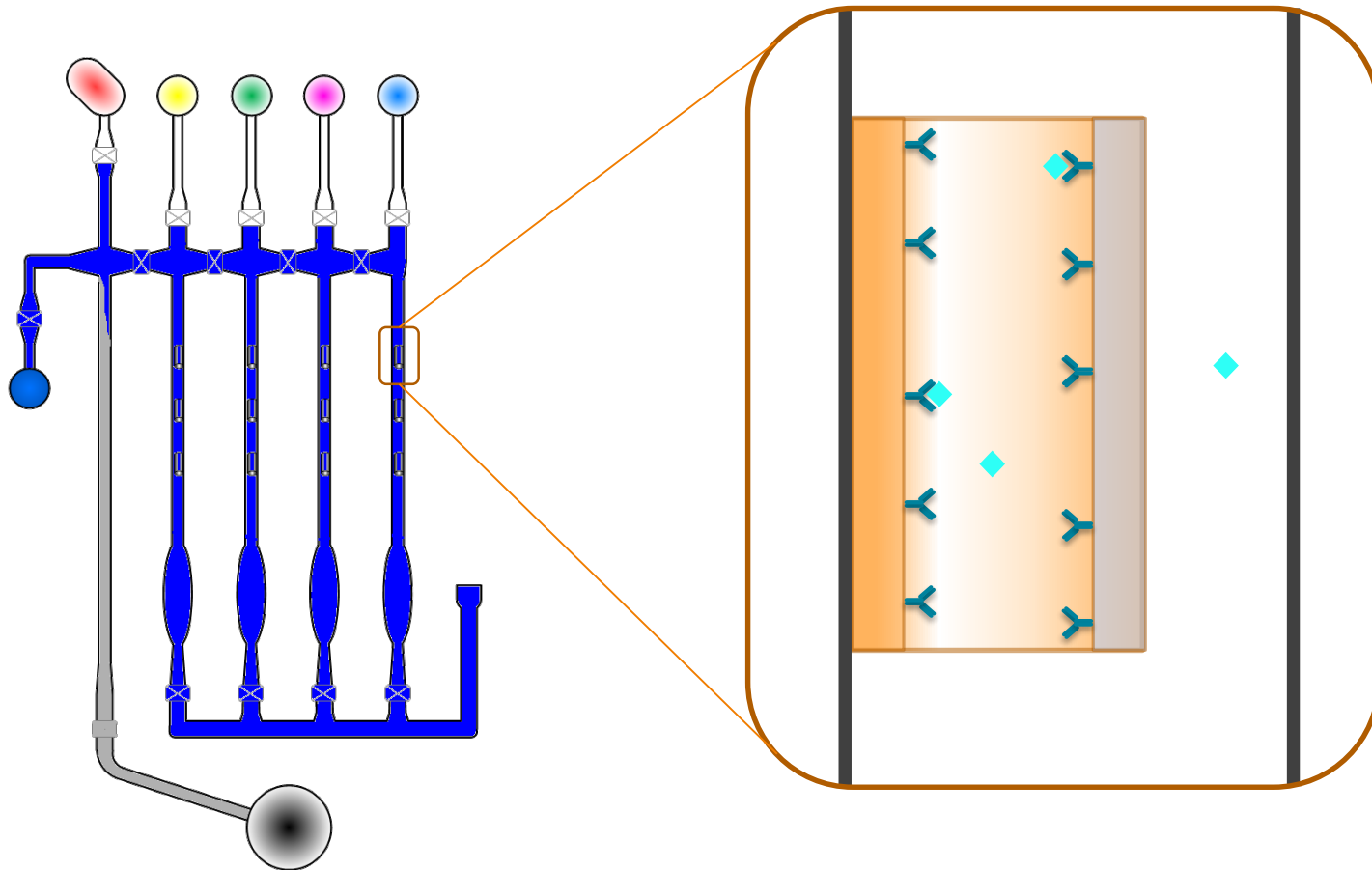
# How Simple Plex works—Sample routing



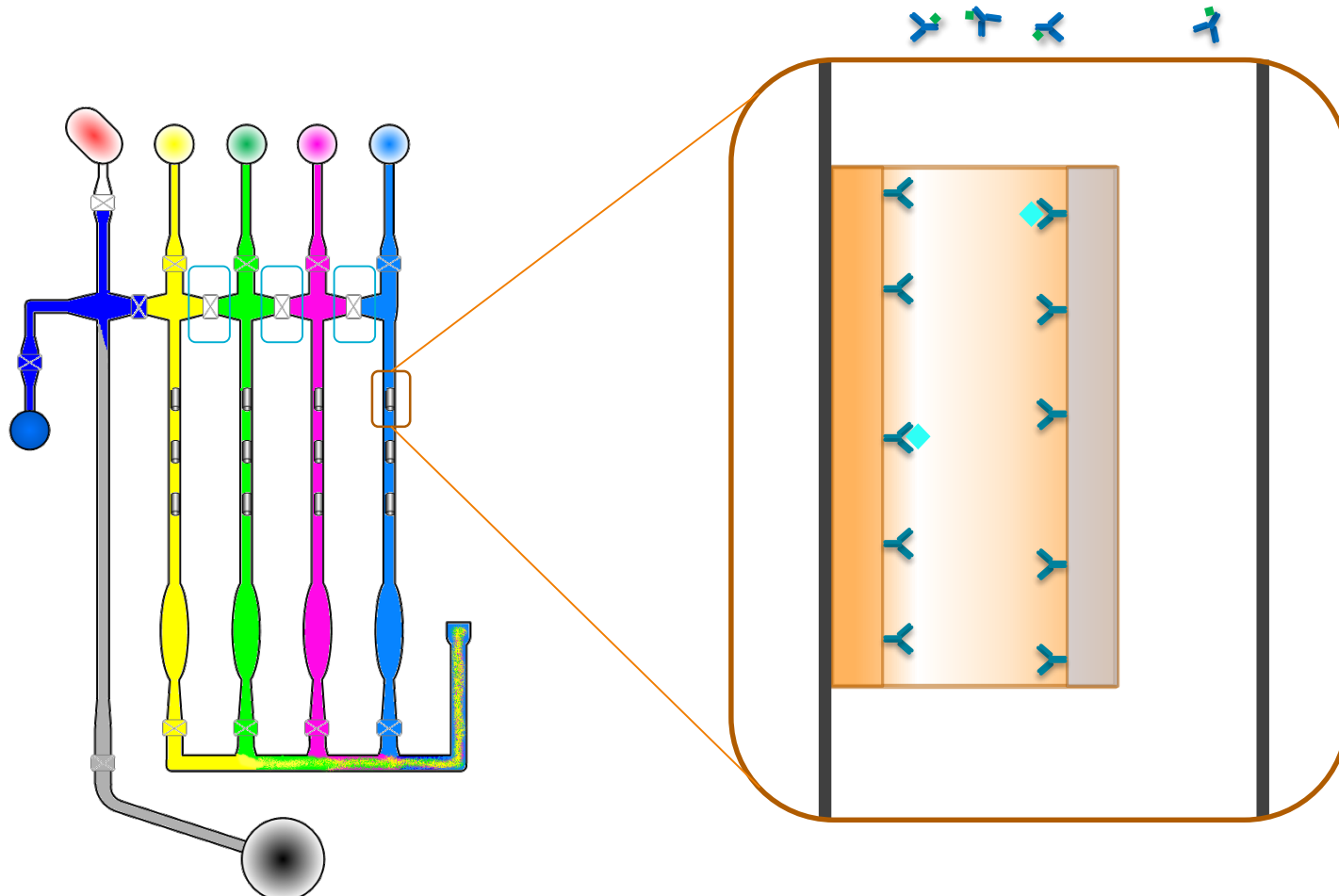
# How Simple Plex works-Sample binding



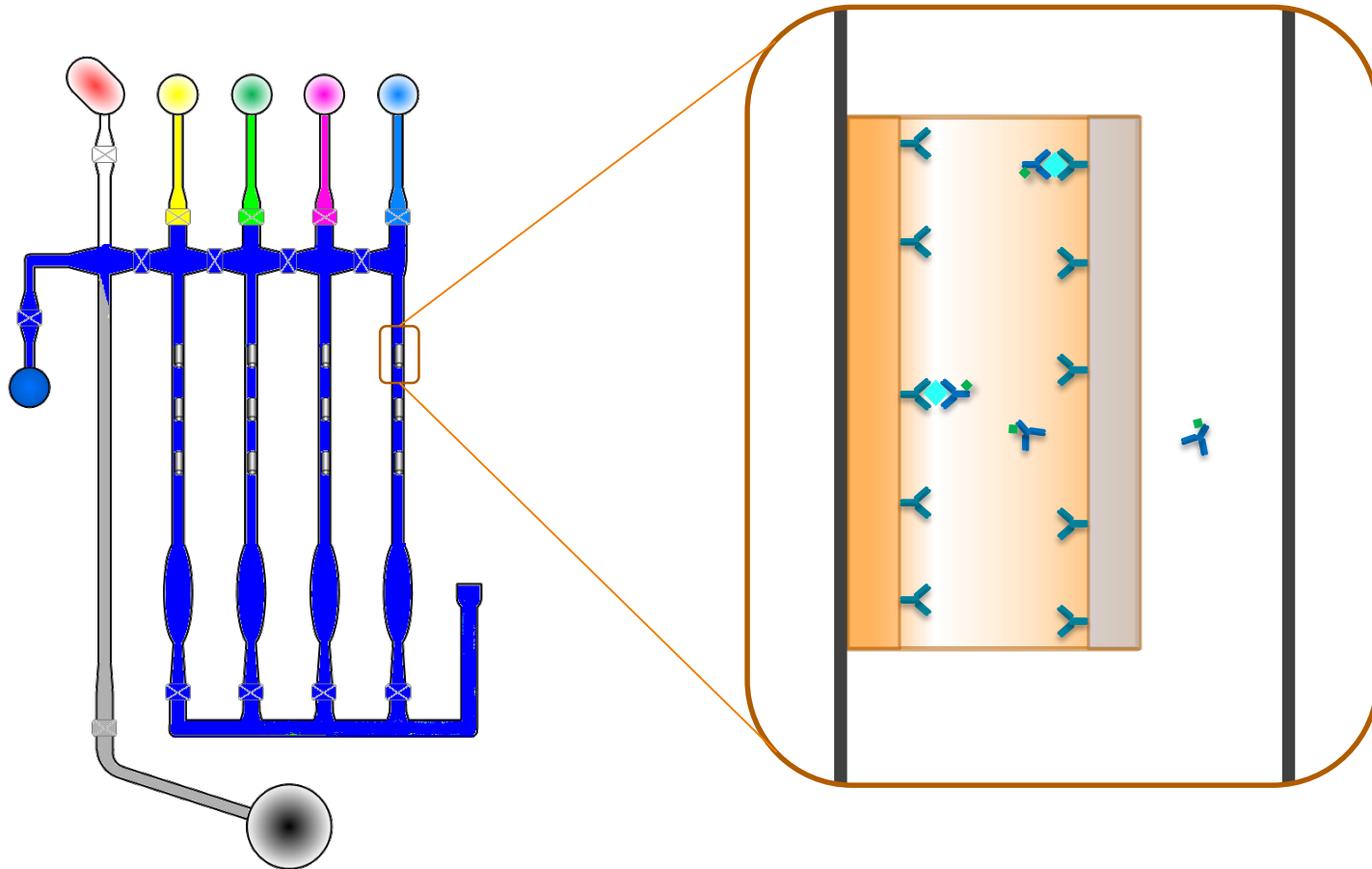
# How Simple Plex works—Wash after sample



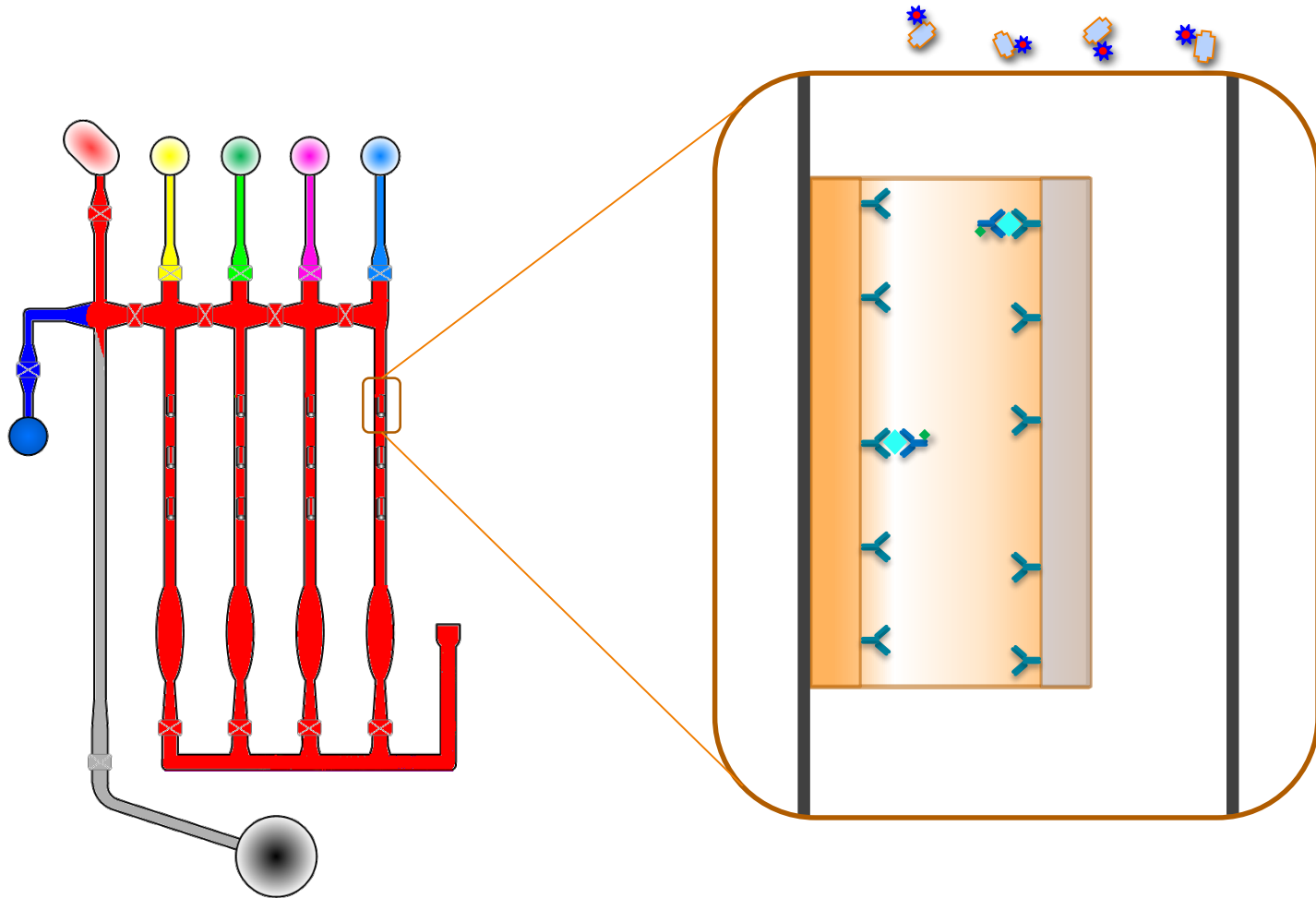
# How Simple Plex works—Antibody addition



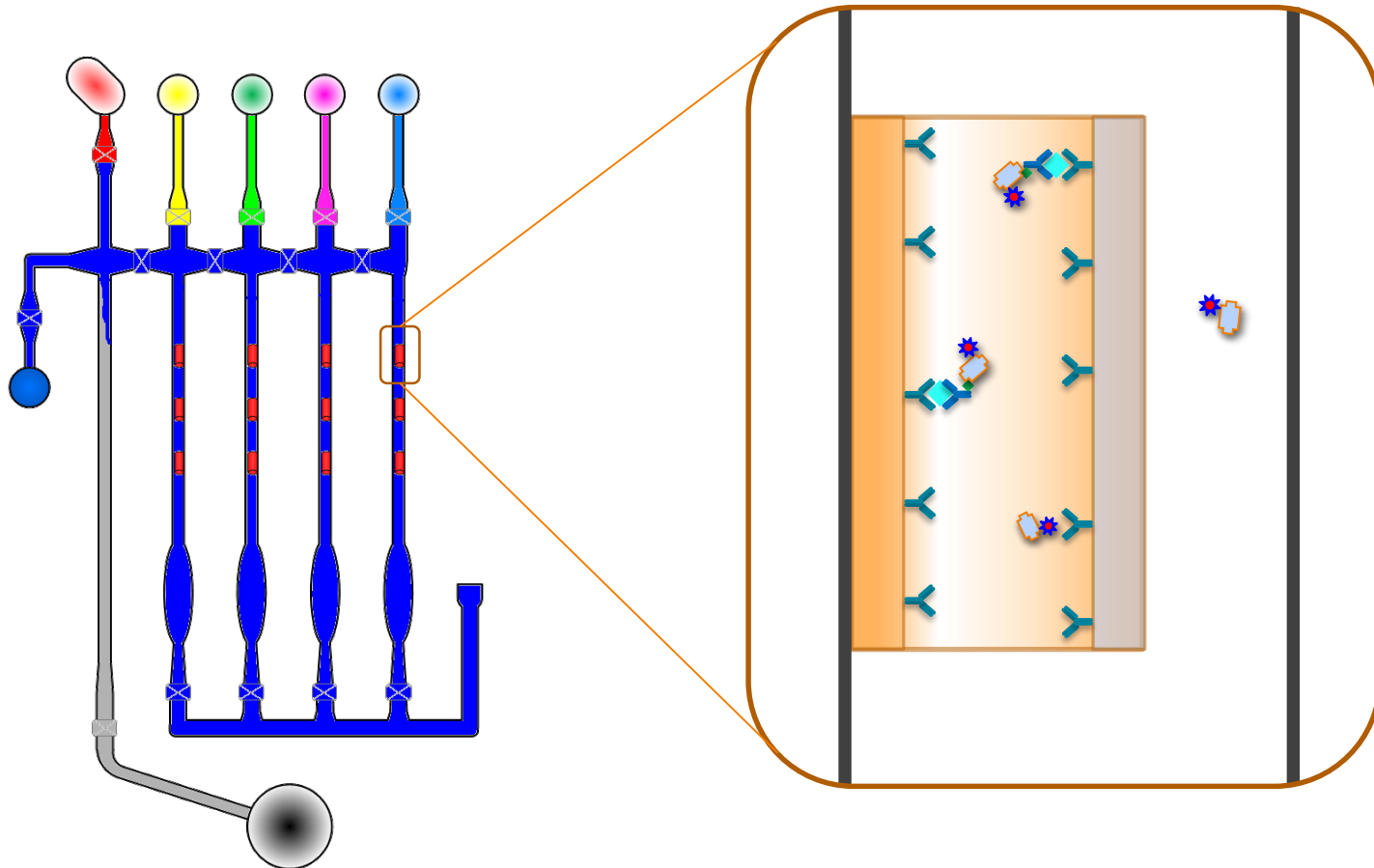
# How Simple Plex works—Wash after sandwich



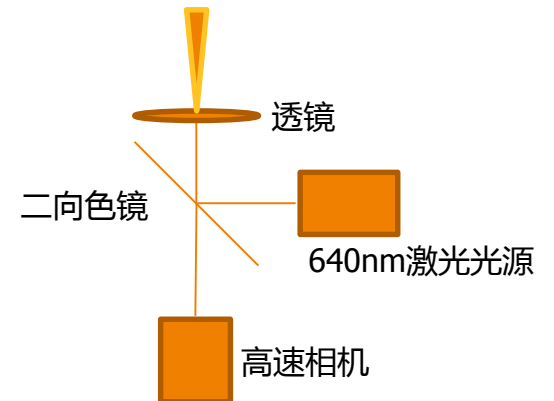
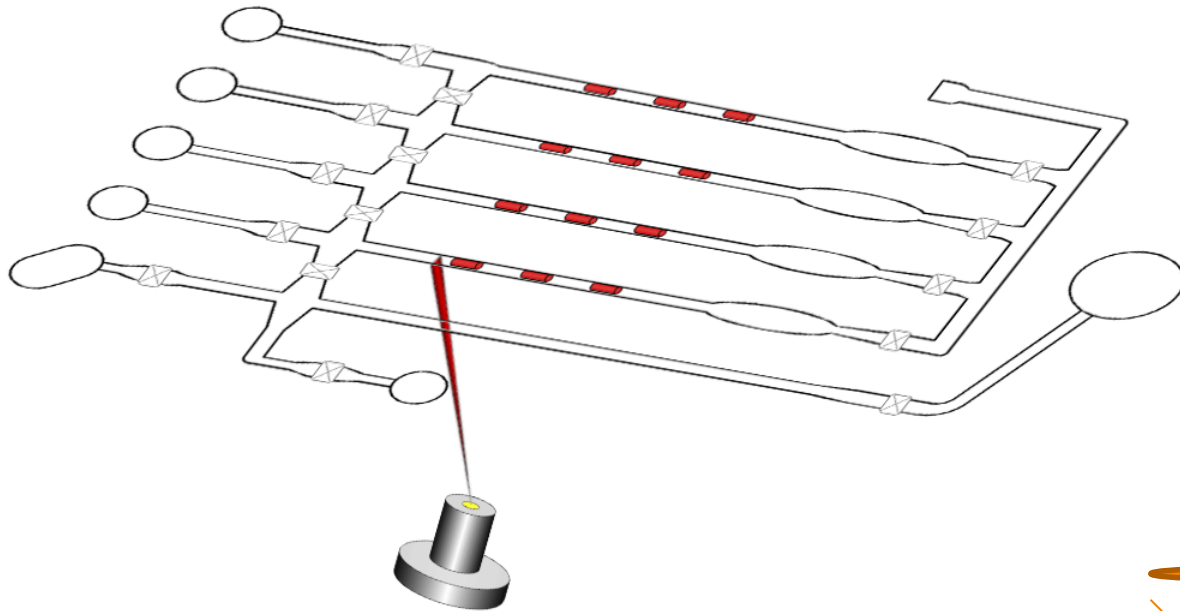
# How Simple Plex works-Dye



# How Simple Plex works—Wash after dye



# How Simple Plex works-Scan



# 功能一：系统自动输出浓度结果, CV < 10%

- 检测结束直接输出浓度结果
- 无需手工进行数据计算分析

### Kit Results

Kit Id: 1000 - Cartridge Id: 9427  
Analytes: IL-1b, IL-6, IL-10, TNF-a  
Comments:

Inlet	Sample	IL-1b					IL-6					GNR
		GNRs	Mean RFU	RFU %CV	Mean Conc....	Conc. %CV	GNRs	Mean RFU	RFU %CV	Mean Conc....	Conc. %CV	
1	serum 1	3	77.15	0.42	<b>126.03</b>	0.50	3	13.05	0.85	<b>56.40</b>	1.02	3
2	serum 2	3	81.19	0.45	<b>133.86</b>	0.53	3	12.80	1.49	<b>55.12</b>	1.78	3
3	serum 3	3	76.45	1.18	<b>124.69</b>	1.39	3	12.64	0.35	<b>54.26</b>	0.42	3
4	serum 4	3	79.35	0.19	<b>130.28</b>	0.23	3	12.85	0.28	<b>55.38</b>	0.33	3
5	serum 5	3	74.27	3.20	<b>120.54</b>	3.76	3	12.15	3.88	<b>51.79</b>	4.65	3
6	serum 6	3	76.87	1.00	<b>125.49</b>	1.18	3	12.34	2.75	<b>52.74</b>	3.29	3
7	serum 7	3	77.32	1.04	<b>126.36</b>	1.22	3	13.03	1.53	<b>56.30</b>	1.83	3
8	low QC	3	79.40	0.46	<b>130.39</b>	0.54	3	12.91	2.05	<b>55.65</b>	2.46	3
9	plasma 1	3	77.31	1.53	<b>126.33</b>	1.81	3	12.71	0.45	<b>54.65</b>	0.54	3
10	plasma 2	3	79.19	0.28	<b>129.98</b>	0.33	3	12.71	1.46	<b>54.64</b>	1.75	3
11	plasma 3	3	74.76	1.28	<b>121.45</b>	1.51	3	12.52	0.62	<b>53.68</b>	0.75	3
12	plasma 4	3	78.36	0.81	<b>128.37</b>	0.96	3	12.36	0.44	<b>52.83</b>	0.53	3
13	plasma 5	3	76.36	0.64	<b>124.51</b>	0.75	3	12.30	1.03	<b>52.55</b>	1.23	3
14	plasma 6	3	76.96	1.01	<b>125.66</b>	1.18	3	12.31	2.54	<b>52.57</b>	3.04	3
15	plasma 7	3	76.06	0.80	<b>123.93</b>	0.94	3	12.40	2.91	<b>53.07</b>	3.48	3
16	high QC	3	73.89	0.87	<b>119.80</b>	1.02	3	12.25	1.00	<b>52.28</b>	1.20	3

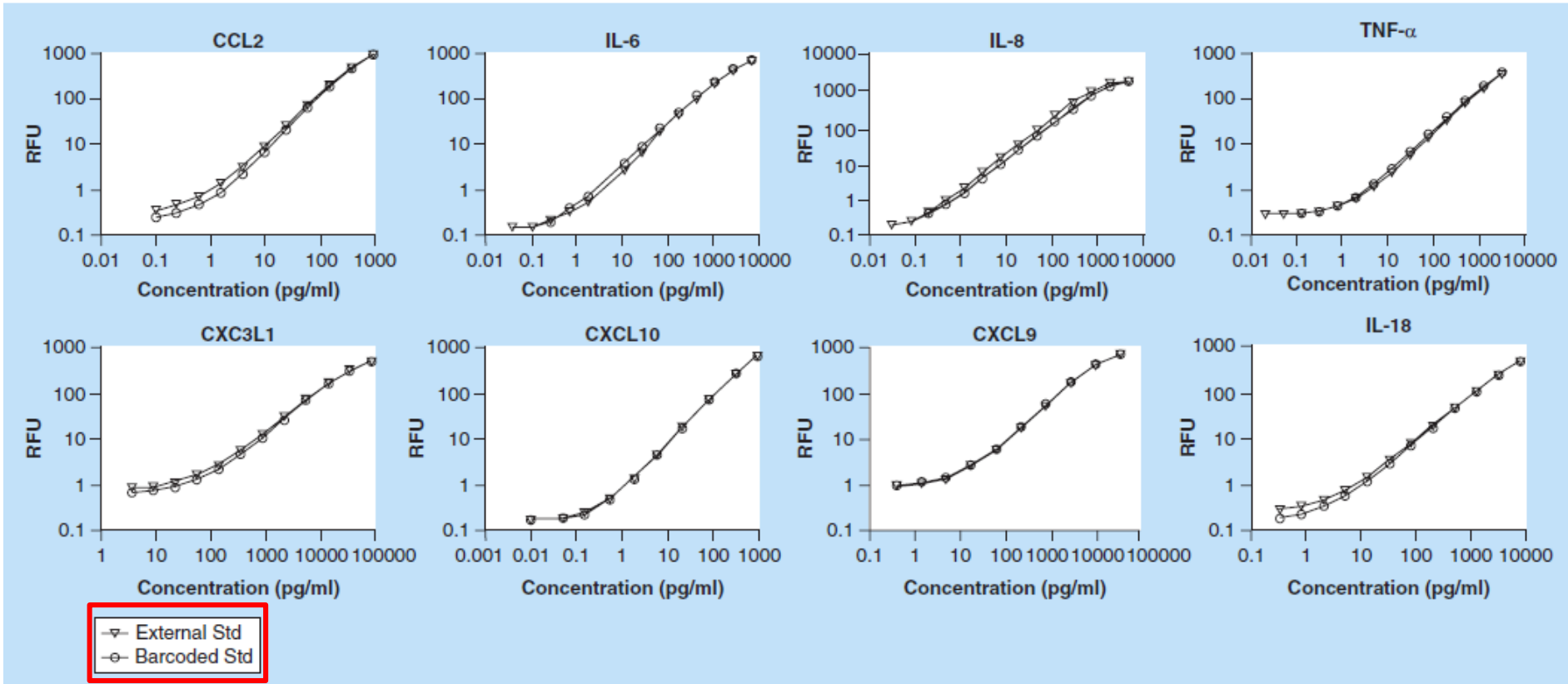
Filter GNRs By: RFU CV Threshold (% CV) 10.0

Save GNR Selection Changes

Kit results saved to Kit001000(4).cvdat in C:\Users\Pu

IL-6				
GNRs	Mean RFU	RFU %CV	Mean Conc....	Conc. %CV
3	13.05	0.85	<b>56.40</b>	1.02
3	12.80	1.49	<b>55.12</b>	1.78
3	12.64	0.35	<b>54.26</b>	0.42
3	12.85	0.28	<b>55.38</b>	0.33
3	12.15	3.88	<b>51.79</b>	4.65
3	12.34	2.75	<b>52.74</b>	3.29
3	13.03	1.53	<b>56.30</b>	1.83
3	12.91	2.05	<b>55.65</b>	2.46
3	12.71	0.45	<b>54.65</b>	0.54
3	12.71	1.46	<b>54.64</b>	1.75
3	12.52	0.62	<b>53.68</b>	0.75
3	12.36	0.44	<b>52.83</b>	0.53
3	12.30	1.03	<b>52.55</b>	1.23
3	12.31	2.54	<b>52.57</b>	3.04
3	12.40	2.91	<b>53.07</b>	3.48
3	12.25	1.00	<b>52.28</b>	1.20

# 功能二: 内置标曲, 避免板与板间标曲差异



每做一批, 抽样测标准曲线, 拟合出二维码的标准曲线

**内置标准曲线与日常实验室标准曲线一致**

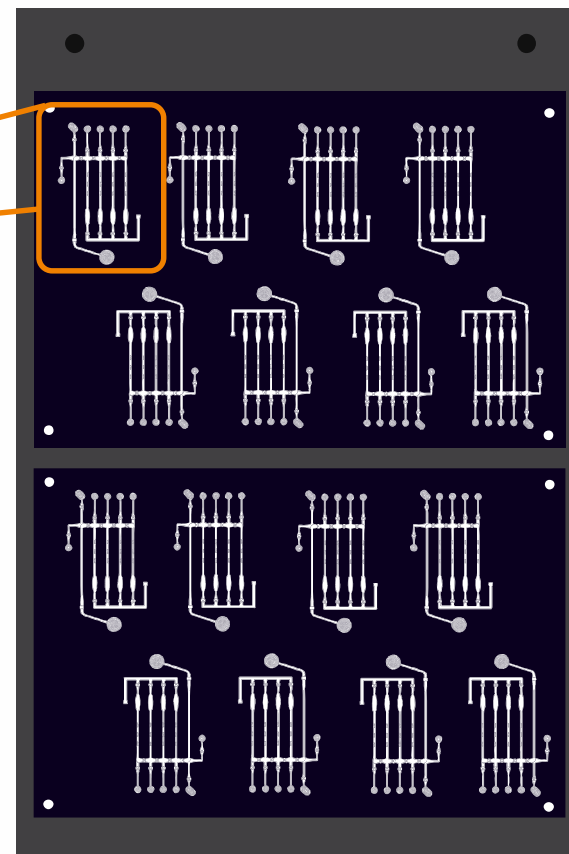
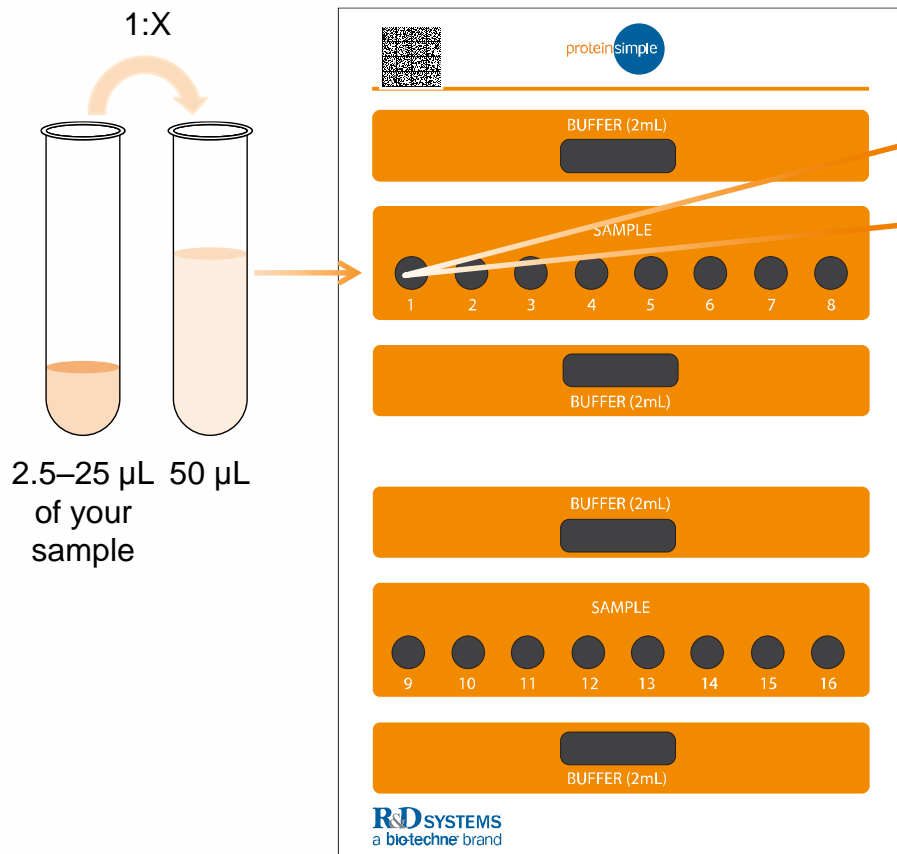
避免每次手工标准曲线差异带来的检测误差 (尤其对于大样本量检测)

Bioanalytical qualification of clinical biomarker assays in plasma using a novel multi-analyte Simple Plex™ platform  
BioAnalytical Sciences, Oncology Biomarker Development **Genentech Inc**, South San Francisco

*Bioanalysis* (2016) 8(23), 2415–2428

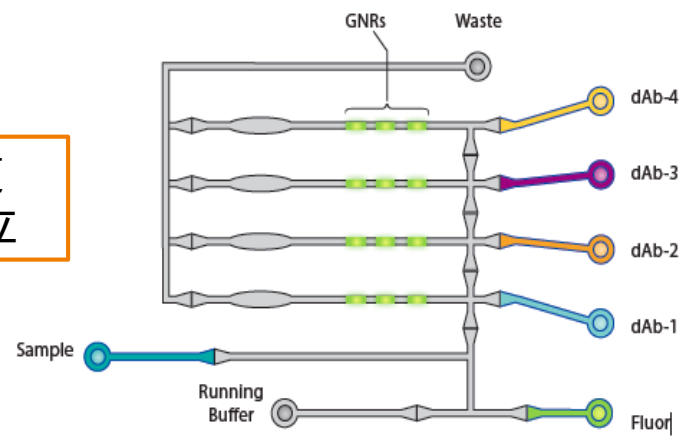
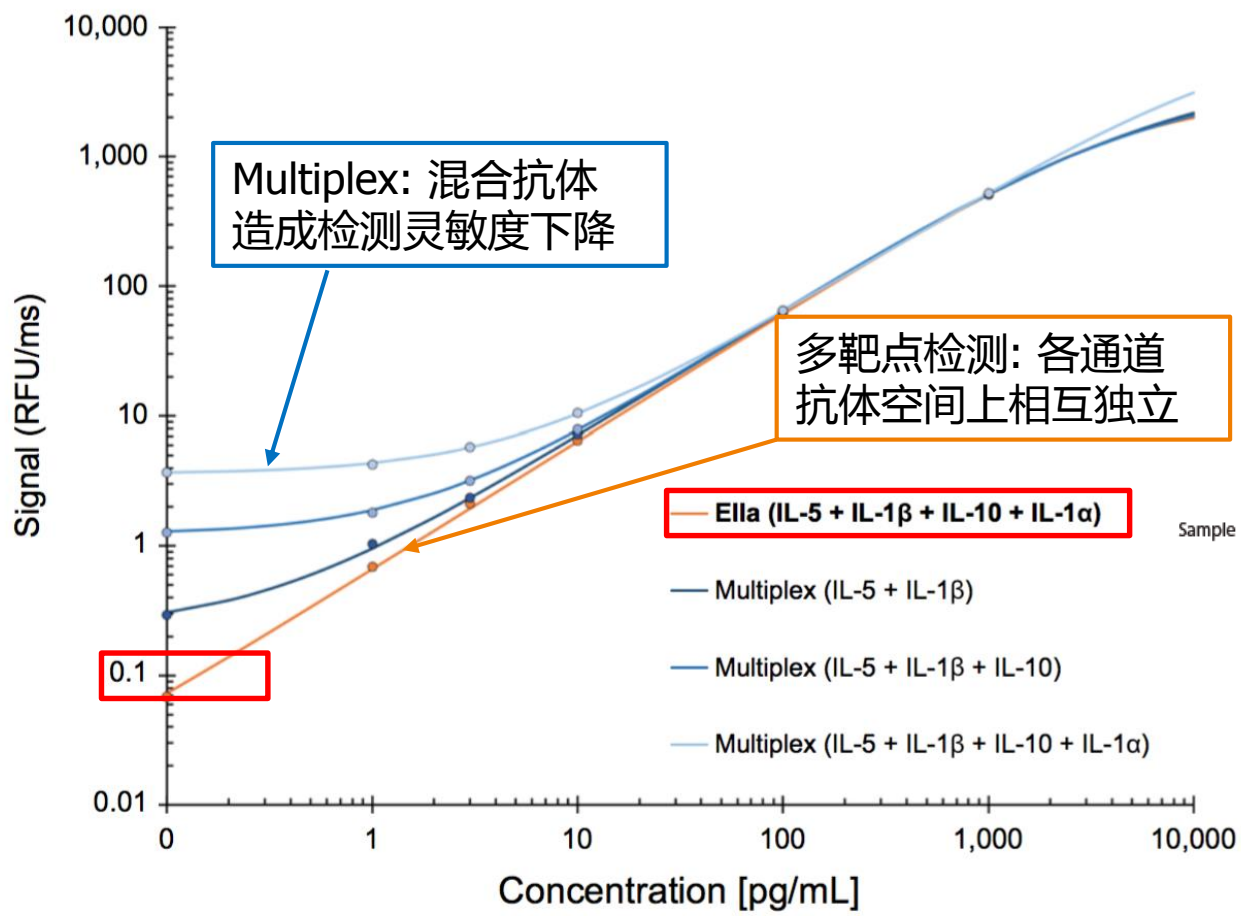


# 功能三:多孔同步吸样,避免孔间反应差



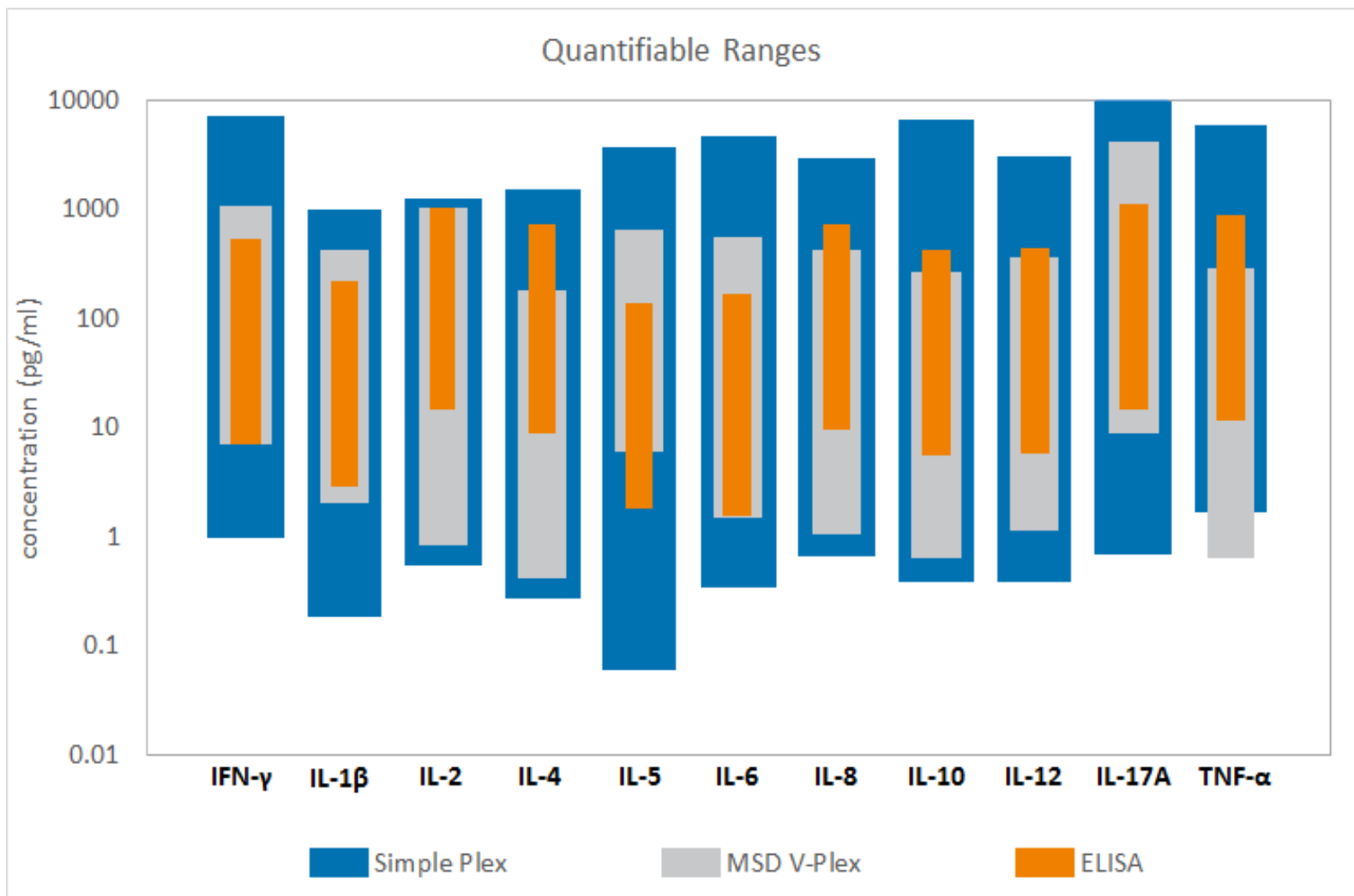
Simple Plex Cartridge 16X4

# 功能四:多因子独立检测,避免抗体交叉污染



**Fig. 3** Effect of multiplexing antibodies to various analytes in the same GNR. The effect of cross-reactivity was determined by adding increasing number of antibodies to the same GNR. Note that the addition of antibodies decreases the sensitivity of the assay.

# 功能五:更宽的检测范围

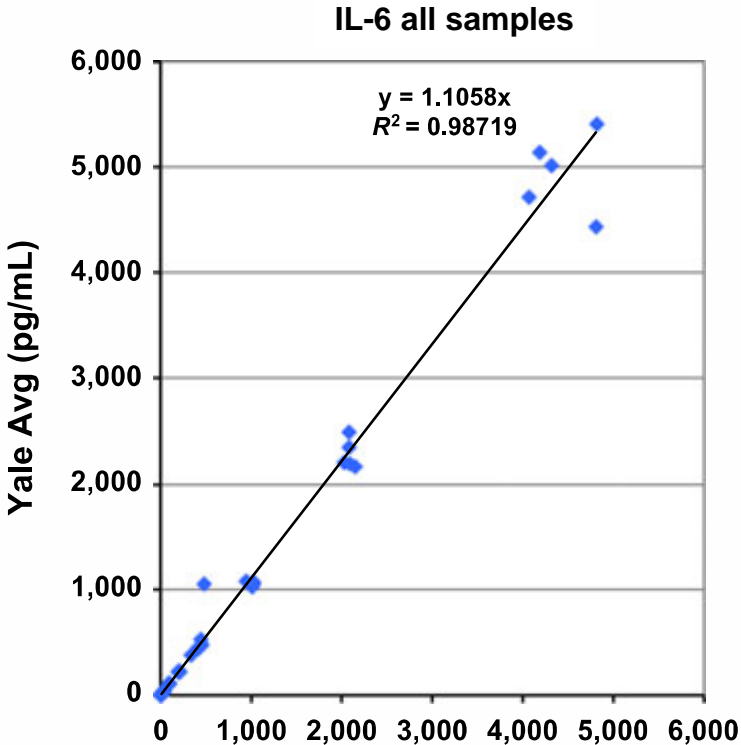
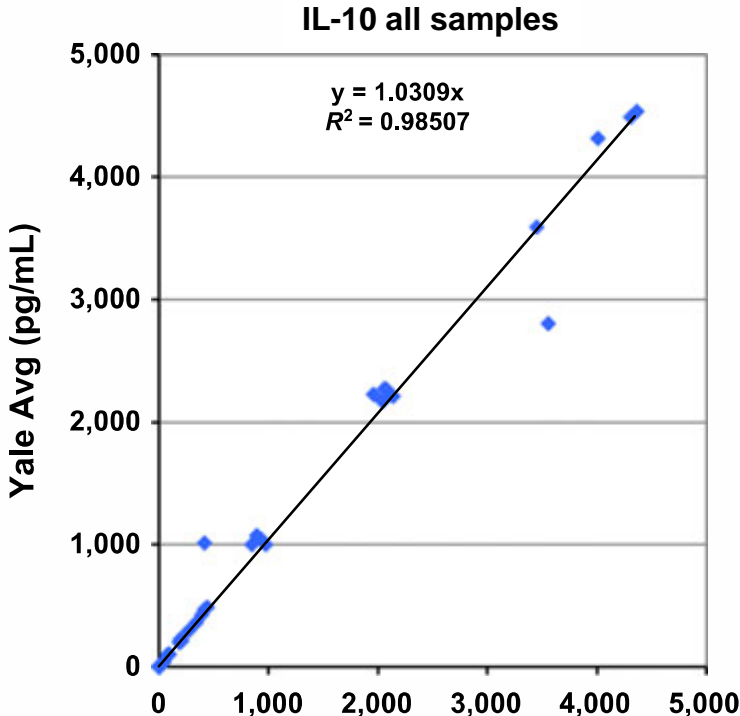


# 功能六: fg及低pg检测灵敏度,只需1h

Parameter	Ella	ELISA	Quantikine	MSD
<b>sensitivity</b>	<b>fg/mL or sub pg/mL</b>	<b>5 pg/mL</b>	<b>1 pg/mL</b>	<b>1pg/mL</b>
dyanmic range	>4 logs	2.5 logs	3 logs	>4 logs
<b>Hands-on time</b>	<b>5 mins</b>	<b>~1.5 h</b>	<b>~1.5 hr</b>	<b>~1.5 h</b>
<b>time to result</b>	<b>1 hr</b>	<b>4- 48 hr</b>	<b>~ 4 hr</b>	<b>4 hr</b>
Sample volume	2.5-25 $\mu$ L	100 $\mu$ L	100 $\mu$ L	75 $\mu$ L
Data points per day	1536	288	288	768

# 功能七: EIIa 极佳的数据重现性

15 个健康人血浆血清样品，2 个不同场所的盲盲试验



◆ IL-10 all samples  
— Linear (IL-10 all samples)

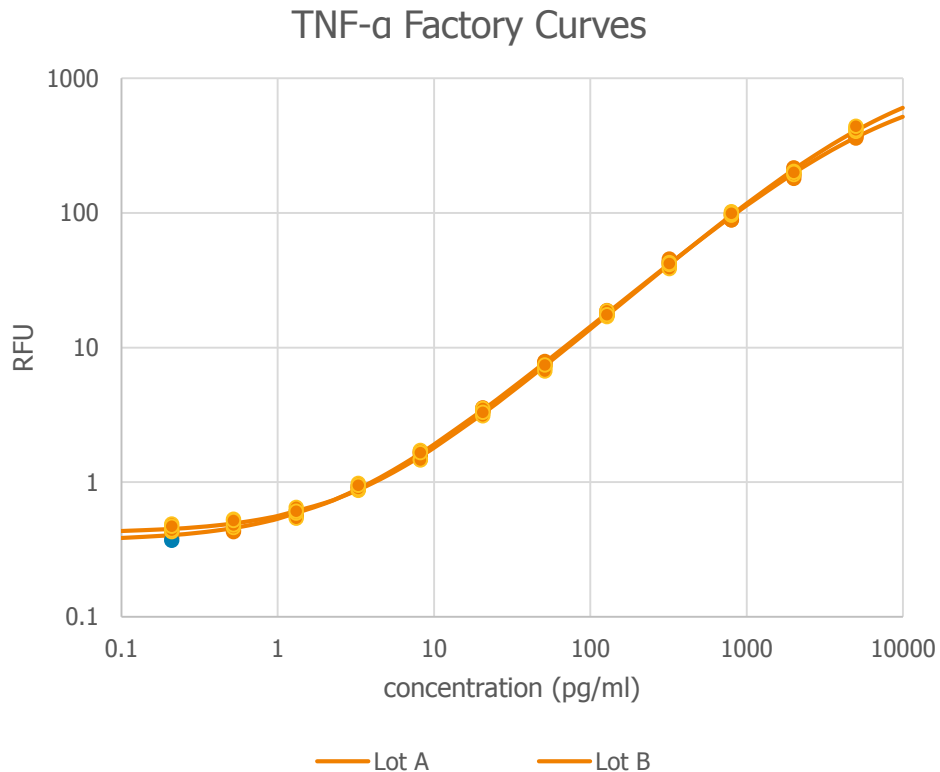
◆ IL-6 all samples  
— Linear (IL-6 all samples)

Plasma/serum samples were run neat and spiked (low, medium, high) for a total of 50 samples.

# 不同天的重复性

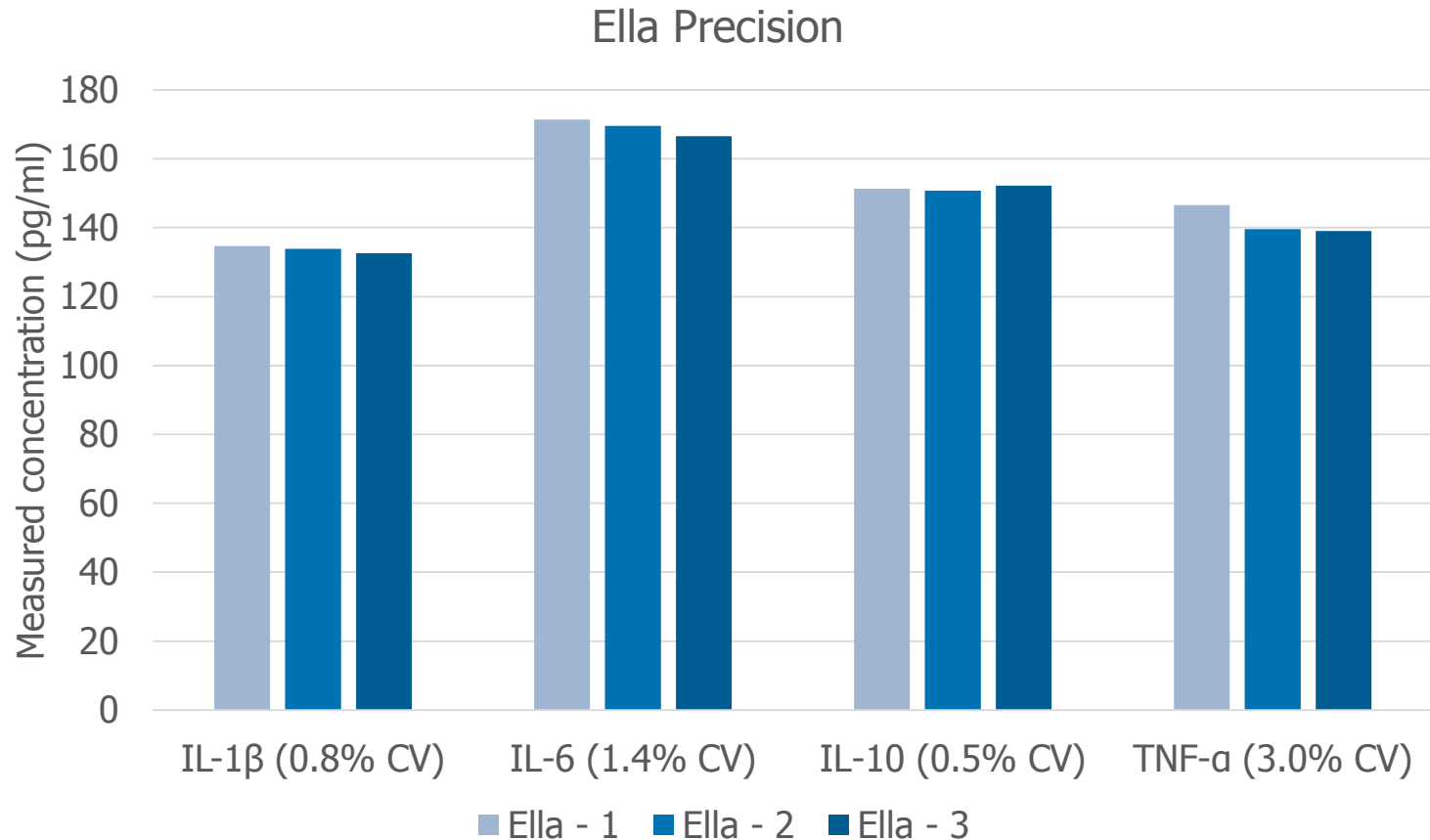
Day #	Control Level 1 (25 pg/ml)				Control Level 2 (500 pg/ml)			
	IL-1 $\beta$	IL-6	IL-10	TNF- $\alpha$	IL-1 $\beta$	IL-6	IL-10	TNF- $\alpha$
1	24.3	24.7	23.2	25.6	504	527	536	541
2	24	24.1	23.8	24.7	576	524	551	511
3	22.6	22.4	23.1	24	539	528	513	467
4	25.2	24.4	23.9	24	674	555	535	484
5	26.5	24.7	24.4	25.9	570	555	535	483
6	27.5	24.7	24.5	26.1	569	573	531	530
7	28.2	24.5	24.3	28	613	575	521	550
8	24.2	24.5	23.1	24.1	530	556	520	479
9	25.4	24.4	24.7	24.2	589	546	523	489
10	24.2	24.2	23.1	23.8	560	536	508	519
Average	25	24	24	25	572	548	527	505
Std Dev	1.73	0.69	0.65	1.35	47.07	18.46	12.75	28.80
CV	7%	3%	3%	5%	8%	3%	2%	6%

# 不同卡盒的重复性

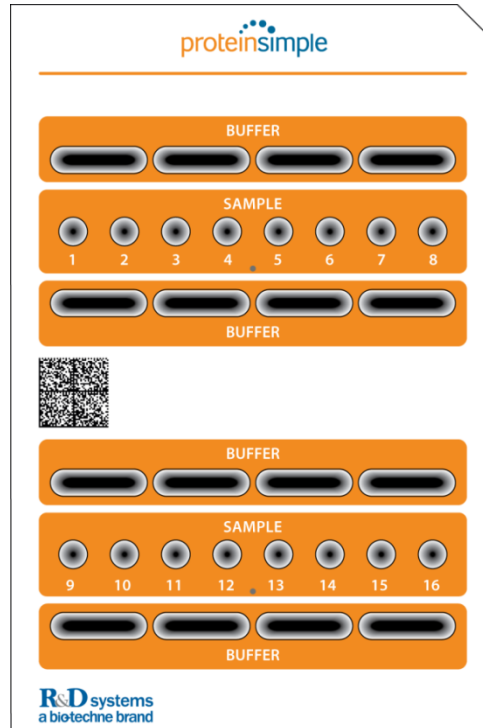
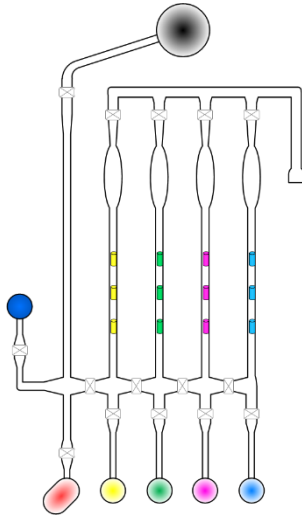


sample	Lot A (pg/ml)	Lot B (pg/ml)	CV
sample - 1	5.601	5.717	1%
sample - 2	6.531	5.887	7%
sample - 3	14.561	16.182	7%
sample - 4	5.58	5.735	2%
sample - 5	6.538	6.474	1%
sample - 6	11.981	12.648	4%
sample - 7	4.744	5.097	5%
sample - 8	3.875	4.163	5%
sample - 9	9.607	10.906	9%
sample - 10	2.7733	3.1339	9%

# 不同仪器的重复性



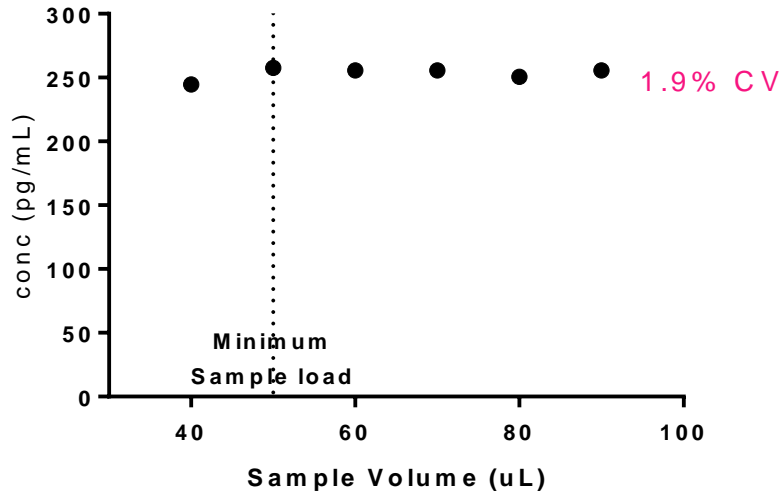
# 不同组件的精密度



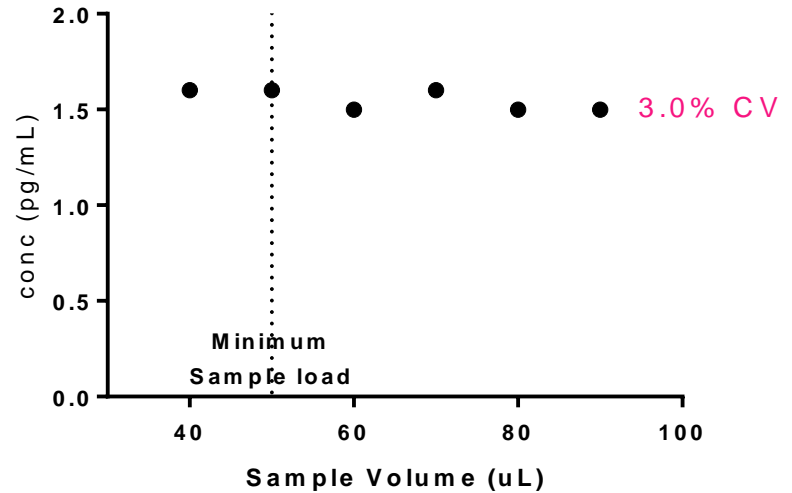
	GNR precision (n=16 channels)	Cartridge precision (n=16 inlets)	Ella precision (n=3 Ella's)
<b>IL-1<math>\beta</math></b>	1%	3%	1%
<b>IL-6</b>	2%	3%	1%
<b>IL-10</b>	2%	4%	1%
<b>TNF-<math>\alpha</math></b>	2%	4%	3%

# 加样体积对结果无显著影响

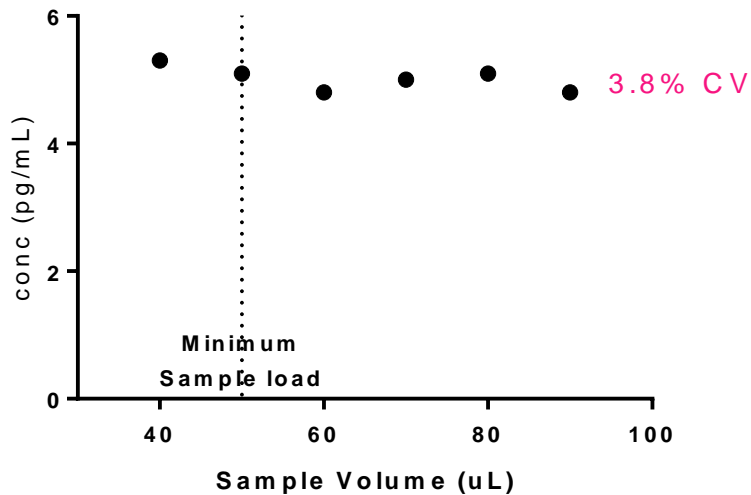
Healthy Plasma Sample - CCL2



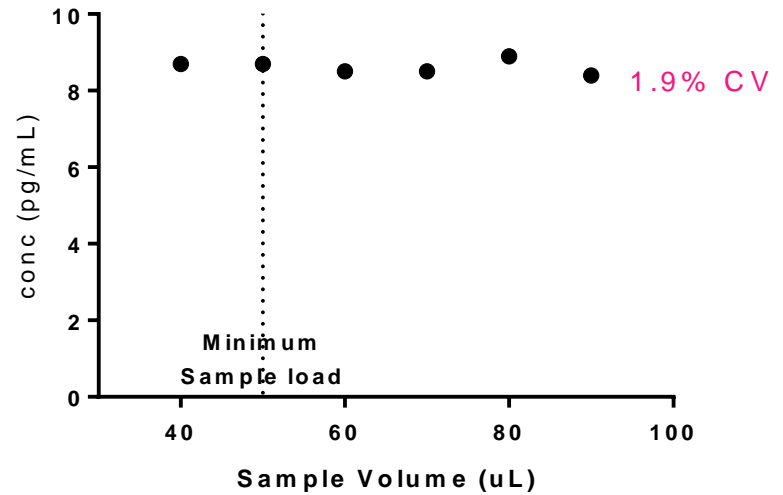
Healthy Plasma Sample - IL-6



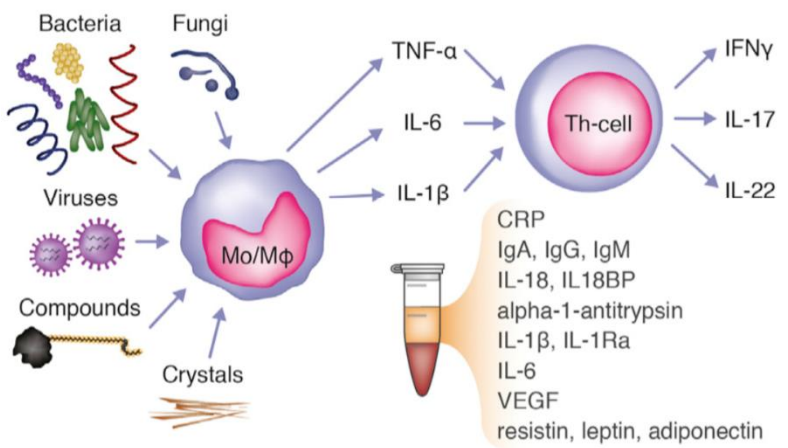
Healthy Plasma Sample - TNF- $\alpha$



Healthy Plasma Sample - VEGF-A



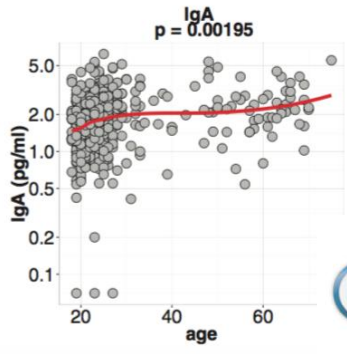
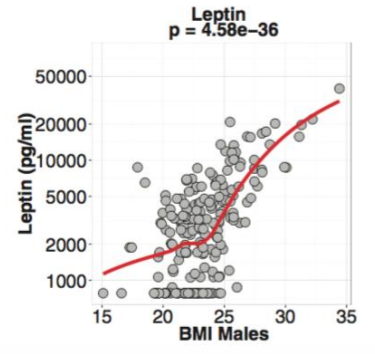
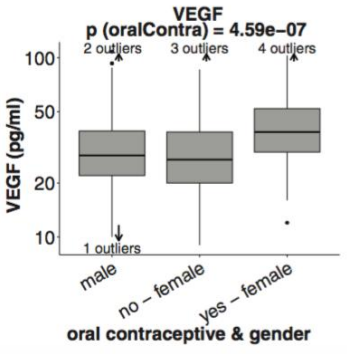
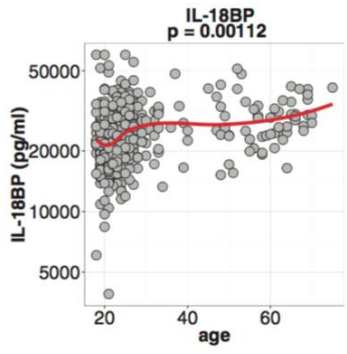
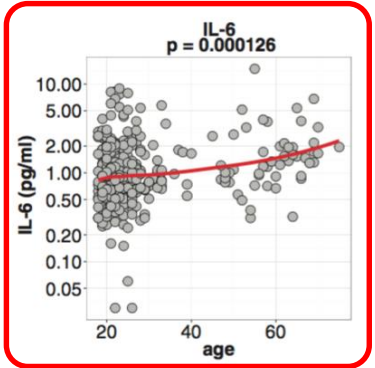
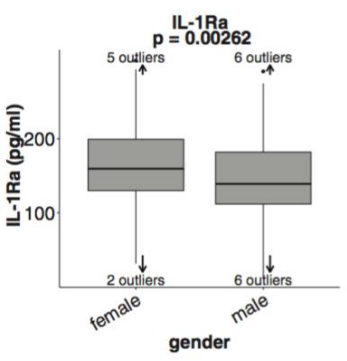
# 功能八: Cell力荐, 超灵敏多因子检测系统



作为最重要的数据: 检测534名健康志愿者细胞因子在静息状态下的表达情况, Ella实现了大规模样品最快速、最灵敏的多因子检测。

### Measurement of Circulating Immune Parameters

To study variation in baseline immune status, we measured circulating concentrations of some of the most important families of parameters responsible for host defense in the circulation: acute phase proteins (CRP and AAT), immunoglobulins (IgA, IgM, IgG, and 4 IgG<sup>+</sup> sub-classes), adipokines (leptin, adiponectin, and resistin), IL-6, IL-18, IL-18-binding protein (IL-18BP), IL-1β, and interleukin-1 receptor antagonist (IL-1Ra). Measurement of resting levels of low abundance cytokines such as IL-1β, IL-6, IL-18, and VEGF are below the lower limit of quantification by standard ELISAs in a healthy cohort; therefore, the Ella microfluidic analyzer was used to assess cytokine concentrations in the fg/mL to low pg/mL range. For instance, the mean level of IL-6 concentrations of the cohort was 1.25 ± 0.06 pg/mL (range



### Host and Environmental Factors Influencing Individual Human Cytokine Responses



ter Horst et al., 2016, Cell 167, 1111-1124  
 November 3, 2016 © 2016 Elsevier Inc.  
<http://dx.doi.org/10.1016/j.cell.2016.10.018>

# 功能九:选择更灵活,三种Kit规格



72样\*单因子\*3复孔

216 Data



32样\*4因子\*3复孔

384 Data



16样\*4因子\*3复孔

192 Data

# 功能十: 广泛, 且不断扩大的抗体库

## Simple Plex Assay Menu (Human)

Acrp30	Enolase-2	IL-17A	Mesothelin	SDF-1α
ANG	EPO	IL-18	MICA	Serpin A4
Ang-1	E-Selectin	IL-18 BPa	MIF	Serpin E1
Ang-2	FABP2	IL-1ra	MIG	SAA
BAFF	FGF-21	IL-1α	MIP-1α	SLPI
BDNF	Follistatin	IL-1β	MIP-1β	TARC
BLC	Fractalkine	IL-2	MMP-1	TIE-2
BMP-2	GAS6	IL-2ra	MMP-7	TIMP-1
BMP-4	GDF-15	IL-3	MMP-8	TNF RI
BMP-9	GH	IL-34	MMP-9	TNF RII
CA125	GM-CSF	IL-4	MPO	TNFSF14
CA9	Granzyme B	IL-5	NCAM-1	TNF-α
CD14	HE4	IL-6	NGAL	TNF-β
CD163	Her2	IL-6Rα	OPN	Trail
CHI3L1	HGF	IL-7	PC9	uPA-1
Tissue factor	ICAM-1	IL-8	PDGF-BB	VCAM-1
CRP	IFN-γ	IP-10	PIGF	VEGF R1
DKK-1	IL-1 R4	LBP	Prolactin	VEGF R2
E-CAD	IL-10	Leptin	RAGE	VEGF-A
EGF	IL-12 p70	MCP-1	Rantes	Vitamin D BP
ENA-78	IL-15	MDC	Reg3A	+4 Mouse Markers

Assay menu is being continually expanded leveraging R&D Systems' portfolio of >600 antibody pairs.

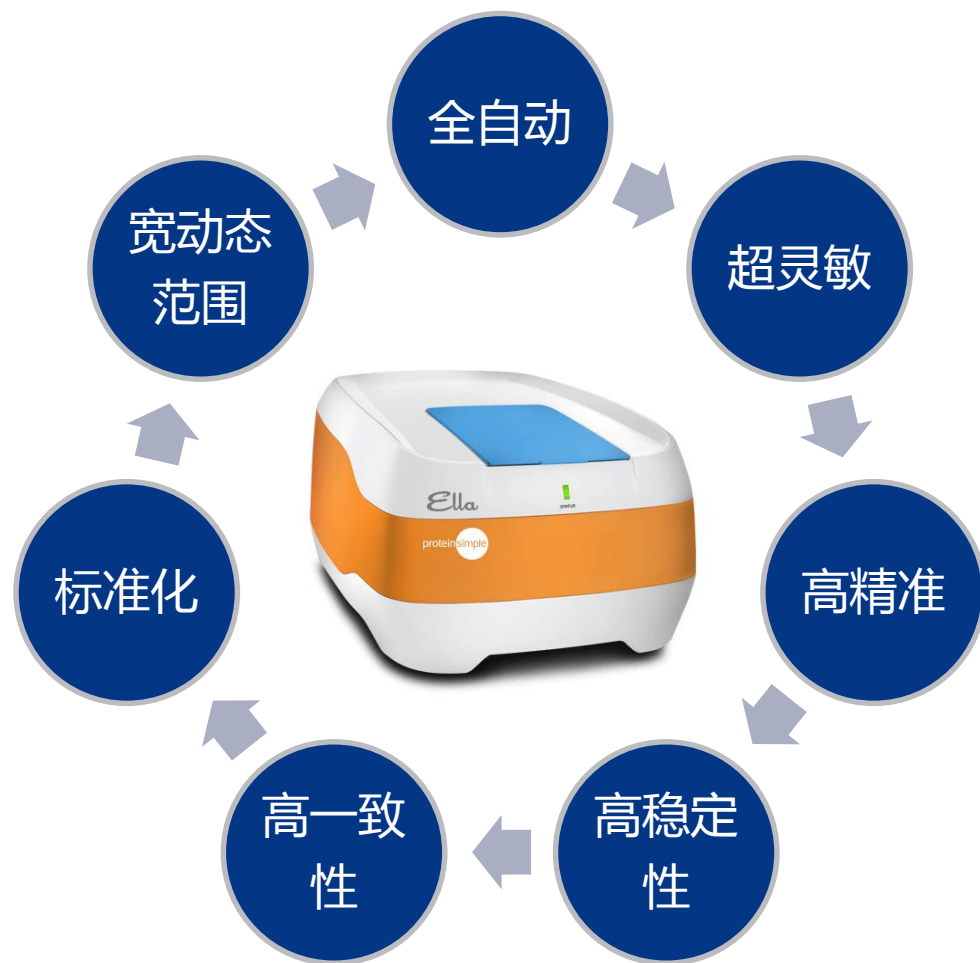
# 功能十一:高度可定制性

## Simple Plex Assay Menu (Human)

Acrp30	Enolase-2	IL-17A	Mesothelin	SDF-1α
ANG	EPO	IL-18	MICA	Serpin A4
Ang-1	E-Selectin	IL-18 BPα	MIF	Serpin E1
Ang-2	FABP2	IL-1ra	MIG	SAA
BAFF	FGF-21	IL-1α	MIP-1α	SLPI
BDNF	Follistatin	IL-1β	MIP-1β	TARC
BLC	Fractalkine	IL-2	MMP-1	TIE-2
BMP-2	GAS6	IL-2ra	MMP-7	TIMP-1
BMP-4	GDF-15	IL-3	MMP-8	TNF RI
BMP-9	GH	IL-34	MMP-9	TNF RII
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E-CAD	IL-10	Leptin	RAGE	VEGF-A
EGF	IL-12 p70	MCP-1	Rantes	Vitamin D BP
ENA-78	IL-15	MDC	Reg3A	+4 Mouse Markers

0.01 0.1 1 10 100 1000 10000  
Concentration (pg/ml)

# Ella 系统特点



谢谢！