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External Quality Assessment Scheme

## Immunohistochemical staining methods Round 3, 2022

### Specimens

Please find enclosed 5 unstained slides S001 - S005.

### Background information

Each participating laboratory receives five unstained slides, one for each staining. Each slide has several sections of paraffin block. The sections of 3-4 µm thickness have been prepared from paraffin embedded tissue fixed in 10% neutral phosphate-buffered formalin (24-48 hours). The sections were cut on slides and have not been dried or otherwise attached to the slides.

### Storage and use

The sections should be dried and attached on slides using heat treatment.

Please store the slides at room temperature before and after staining. If all the mentioned antibodies are not included in your selection, you can participate also with selected antibodies according to your order: product 6600 for 1-5 antibodies, product 6600S for 1-3 antibodies. Please note that we will send all the 5 slides in both products. If you have ordered 6600S, please return maximum 3 stained slides. If needed, extra slides can be ordered from Labquality.

Please, remember to label each slide clearly with given Labquality account number. If you order multiple sample sets with the same client code, please separate them with roman numerals or the alphabet.

### Returning slides and protocol reporting

Please return the stained slides in the same slide mailer box wrapped in bubble wrap at participant's own cost to the address below unless our local partner has instructed otherwise. An address label is included with your shipment for return.

The slides should be at Labquality's office no later than the date given in the column on the right side of the page. Please note that slides received after that will no longer be included in the evaluation.

For each antibody staining, please fill a separate staining techniques protocol form of the used method via LabScala. We can provide a more comprehensive final report that serves the customer if the method information is given as detailed as possible.

### Examination

S001: CK7 (pancreas, liver, pancreatic adenocarcinoma)  
S002: WT1 (kidney, fallopian tube, omentum: metastasis)  
S003: CK20 (colon, colon adenocarcinoma)  
S004: CDX2 (colon, colon adenocarcinoma)  
S005: PSA (prostate, prostatic adenocarcinoma)

2022-11-01

### INSTRUCTIONS

Product no. 6600 / 6600S  
LQ778322031-035/FI

If the kit is incomplete or contains damaged specimens, please report immediately to [info@labquality.fi](mailto:info@labquality.fi)

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The slides should be returned no later than  
**December 13, 2022.**

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### Inquiries

EQA Coordinator  
Pia Eloranta  
[pia.eloranta@labquality.fi](mailto:pia.eloranta@labquality.fi)

### Labquality Oy

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Finland

Tel. + 358 9 8566 8200  
Fax + 358 9 8566 8280

[info@labquality.fi](mailto:info@labquality.fi)  
[www.labquality.com](http://www.labquality.com)

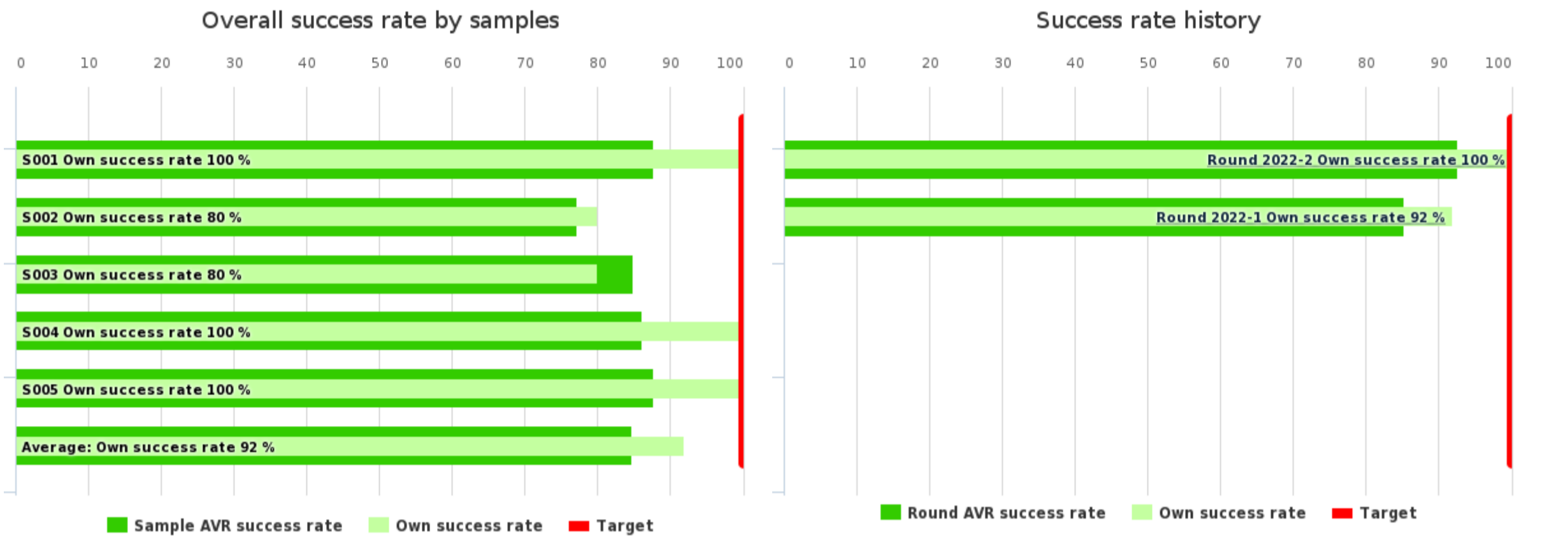
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Client report

	No of participants	No of responded participants	Response percentage
Immunohistochemical staining methods, limited selection of antibodies, November, 3-2022	29	29	100 %
Immunohistochemical staining methods, November, 3-2022	65	65	100 %

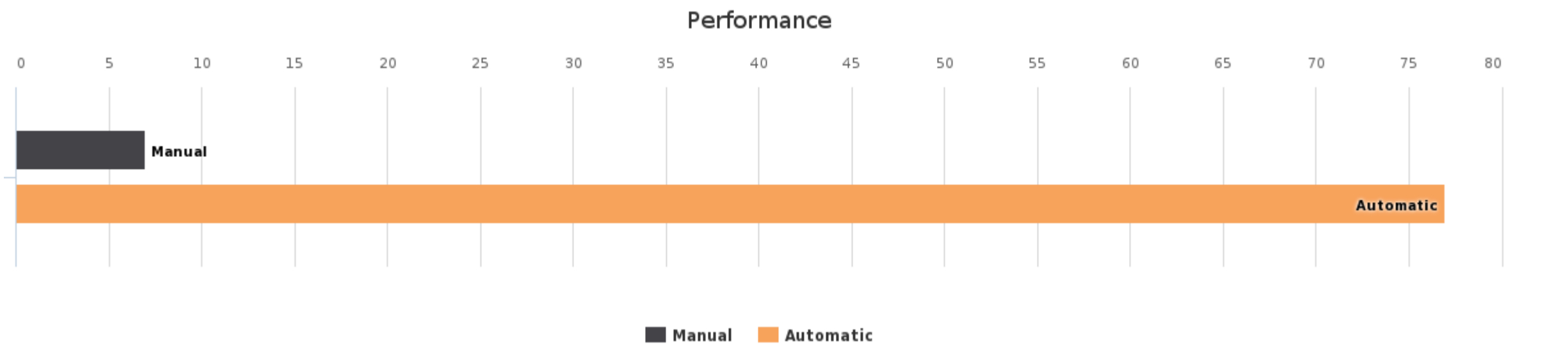
Summary



Summary	Own score	Max score	Own success rate	Difference	AVR success rate
S001	5	5	100 %	12.3 %	87.7 %
S002	4	5	80 %	2.8 %	77.2 %
S003	4	5	80 %	-5 %	85 %
S004	5	5	100 %	13.8 %	86.2 %
S005	5	5	100 %	12.3 %	87.7 %
Average:			92 %	7.2 %	84.8 %

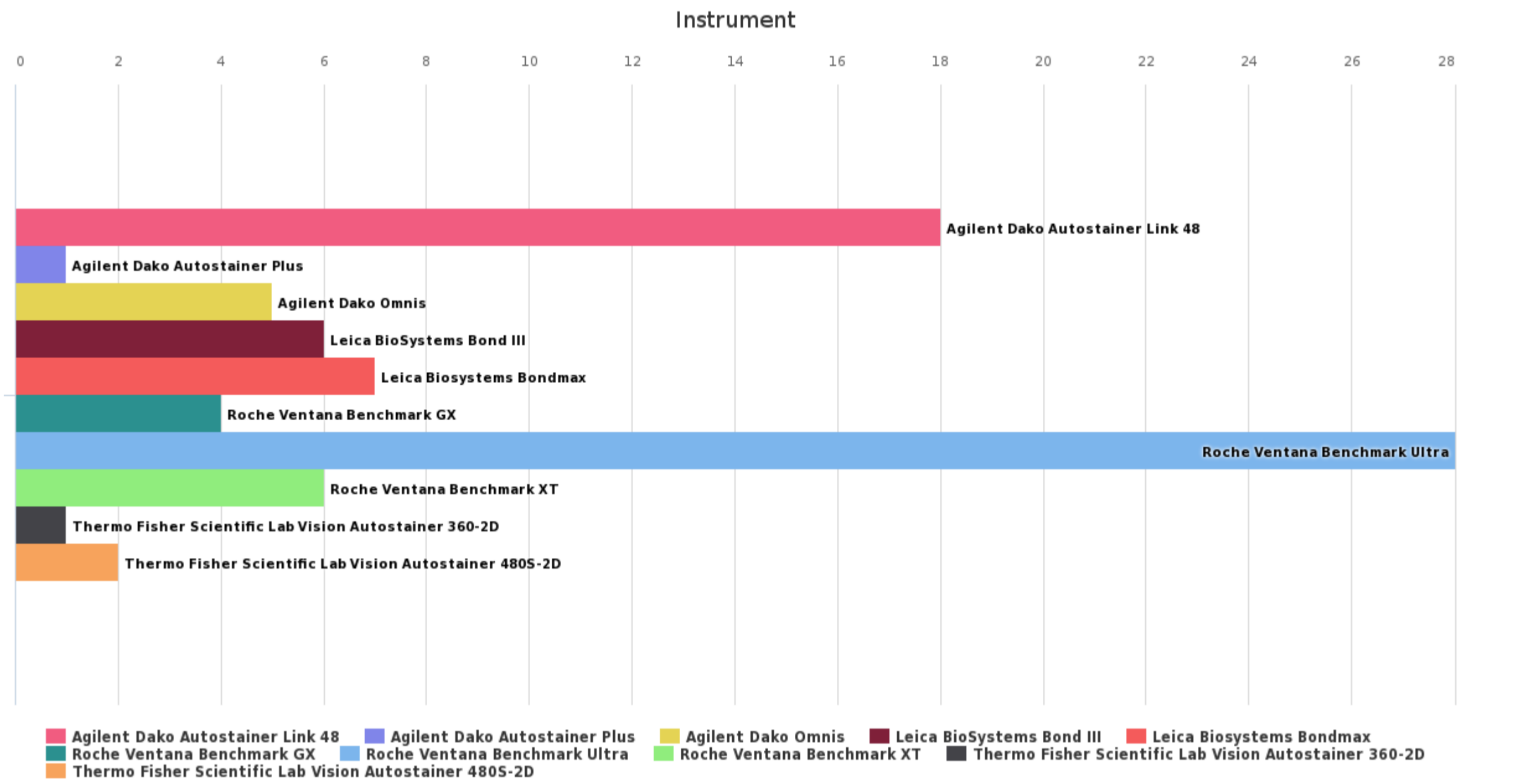
History	Test nr.	Own success rate	Difference	AVR success rate
Round 2022-2	1-1	100 %	7.3 %	92.7 %
Round 2022-1	1-1	92 %	6.7 %	85.3 %

S001 | Sample S001 CK7

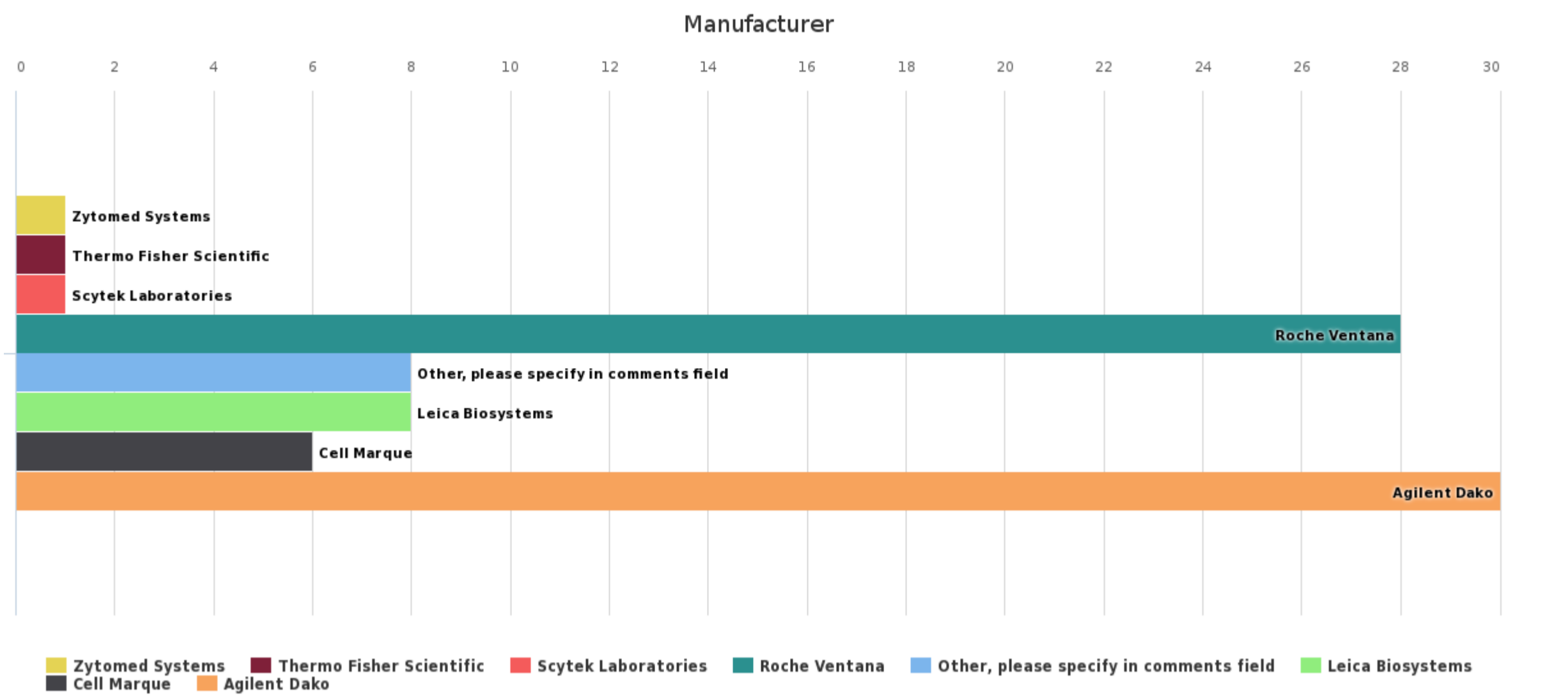


GENERAL DETAILS OF STAINING

Performance	Performance count
<input checked="" type="radio"/> Automatic	77
Manual	7
Total:	84

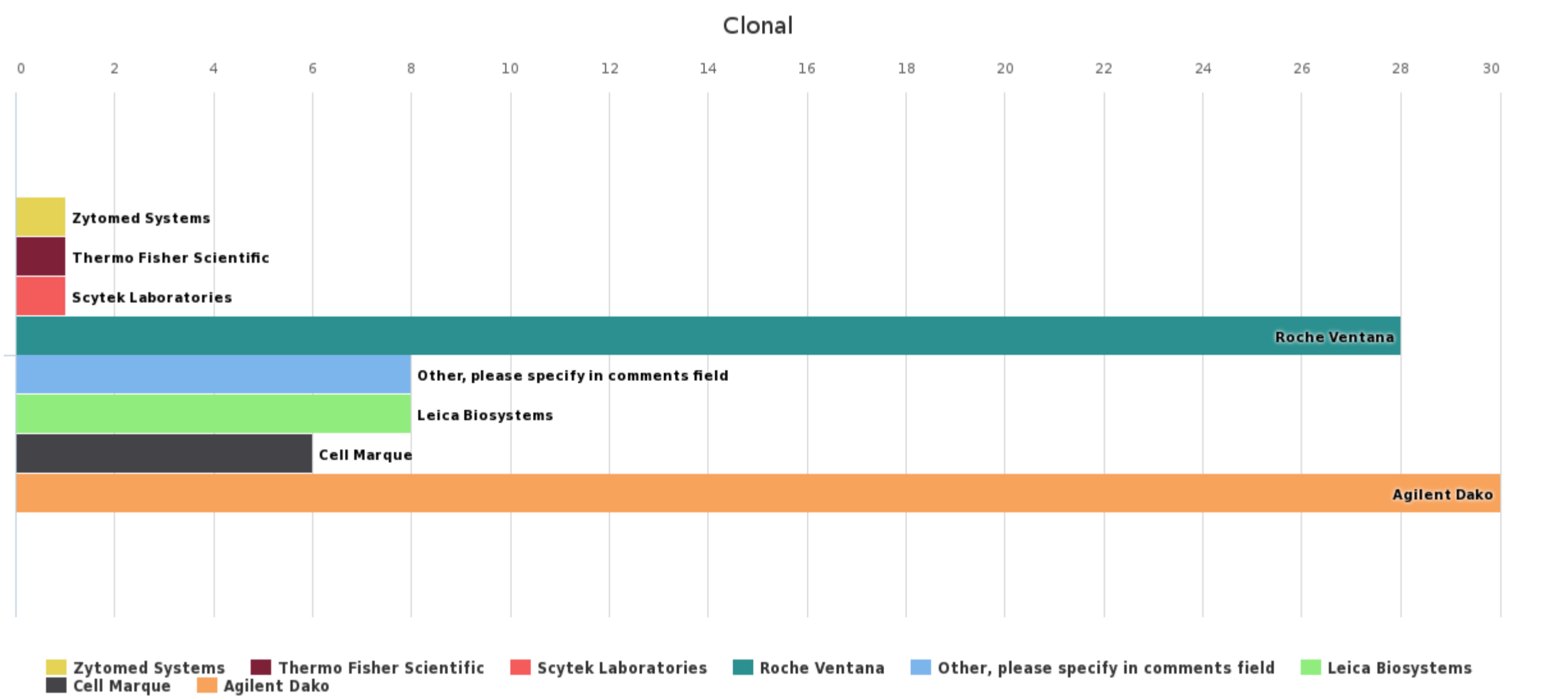


Instrument	Instrument count
Agilent Dako Autostainer Link 48	18
Agilent Dako Autostainer Plus	1
Agilent Dako Omnis	5
Leica BioSystems Bond III	6
Leica Biosystems Bondmax	7
Roche Ventana Benchmark GX	4
<input checked="" type="radio"/> Roche Ventana Benchmark Ultra	28
Roche Ventana Benchmark XT	6
Thermo Fisher Scientific Lab Vision Autostainer 360-2D	1
Thermo Fisher Scientific Lab Vision Autostainer 480S-2D	2
Total:	78

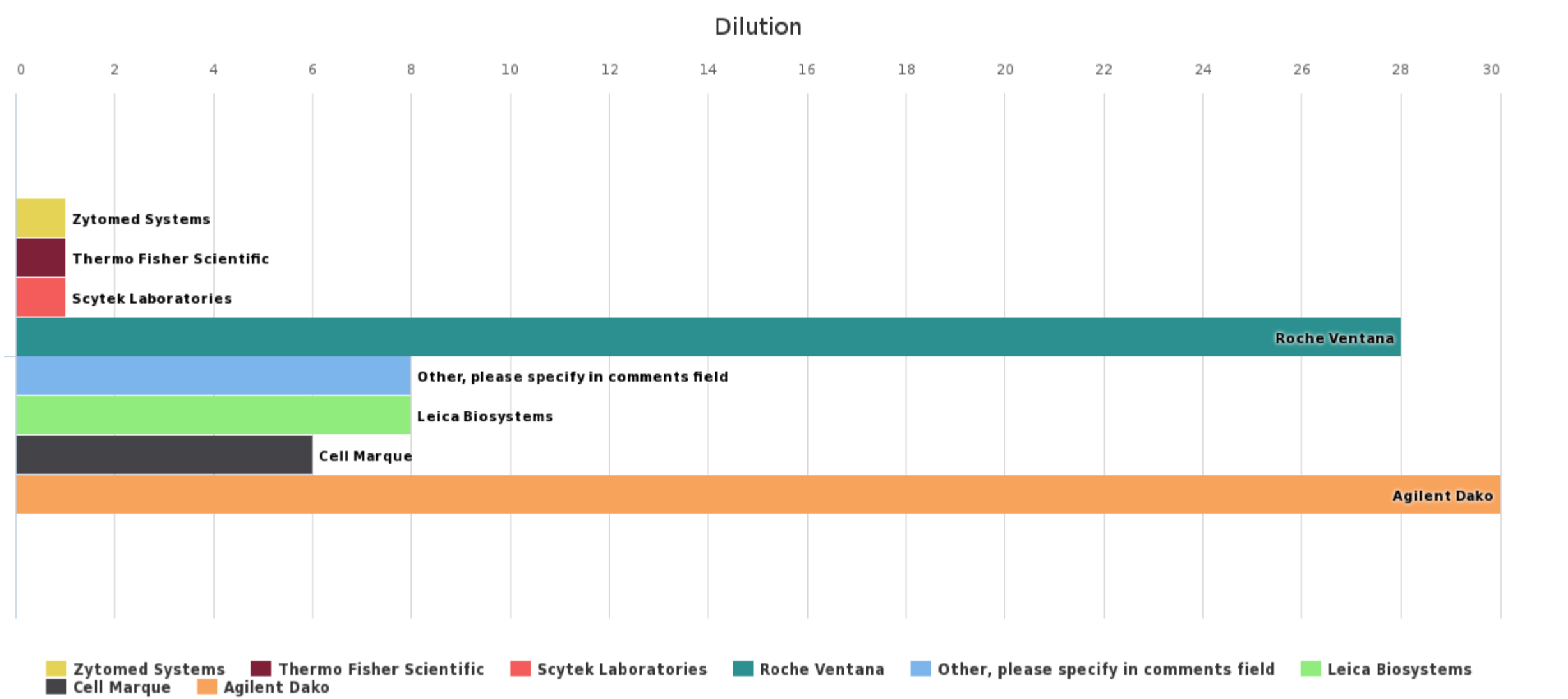


PRIMARY ANTIBODY

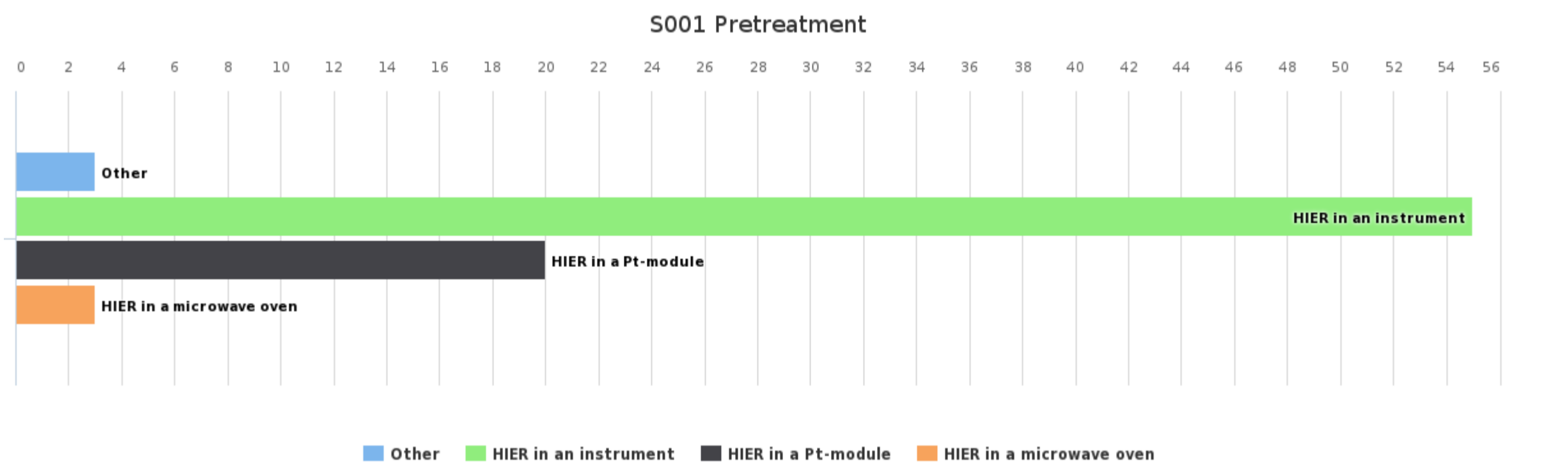
Manufacturer	Manufacturer count
Agilent Dako	30
Cell Marque	6
Leica Biosystems	8
Other, please specify in comments field	8
<input checked="" type="radio"/> Roche Ventana	28
Scytek Laboratories	1
Thermo Fisher Scientific	1
Zytomed Systems	1
Total:	83



Clonal	Clonal count
<input checked="" type="radio"/> Monoclonal	82
Total:	82

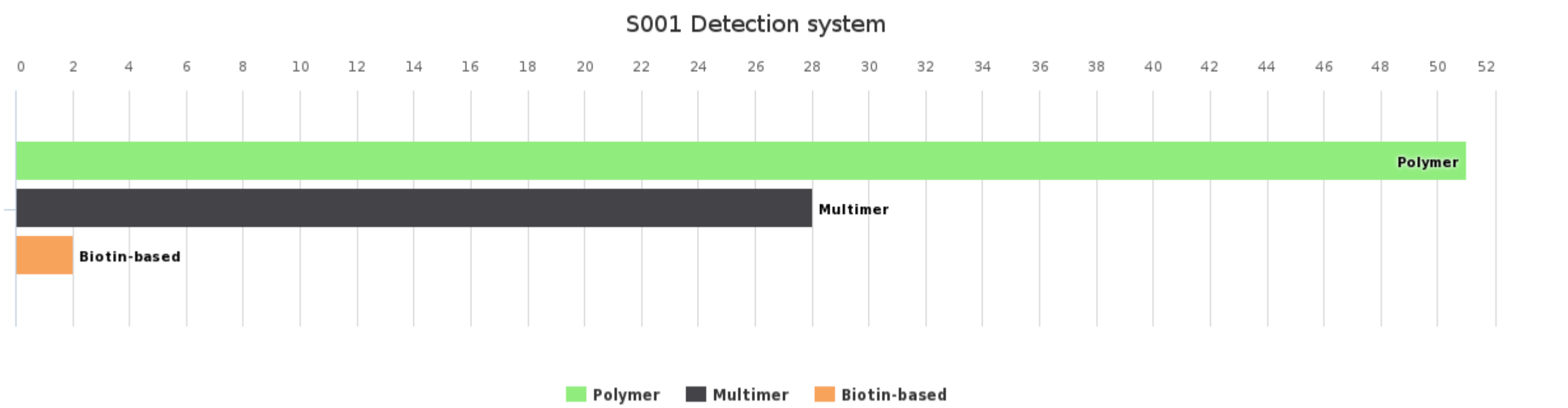


Dilution	Dilution count
Dilution	33
<input checked="" type="radio"/> Ready-to-use	50
Total:	83



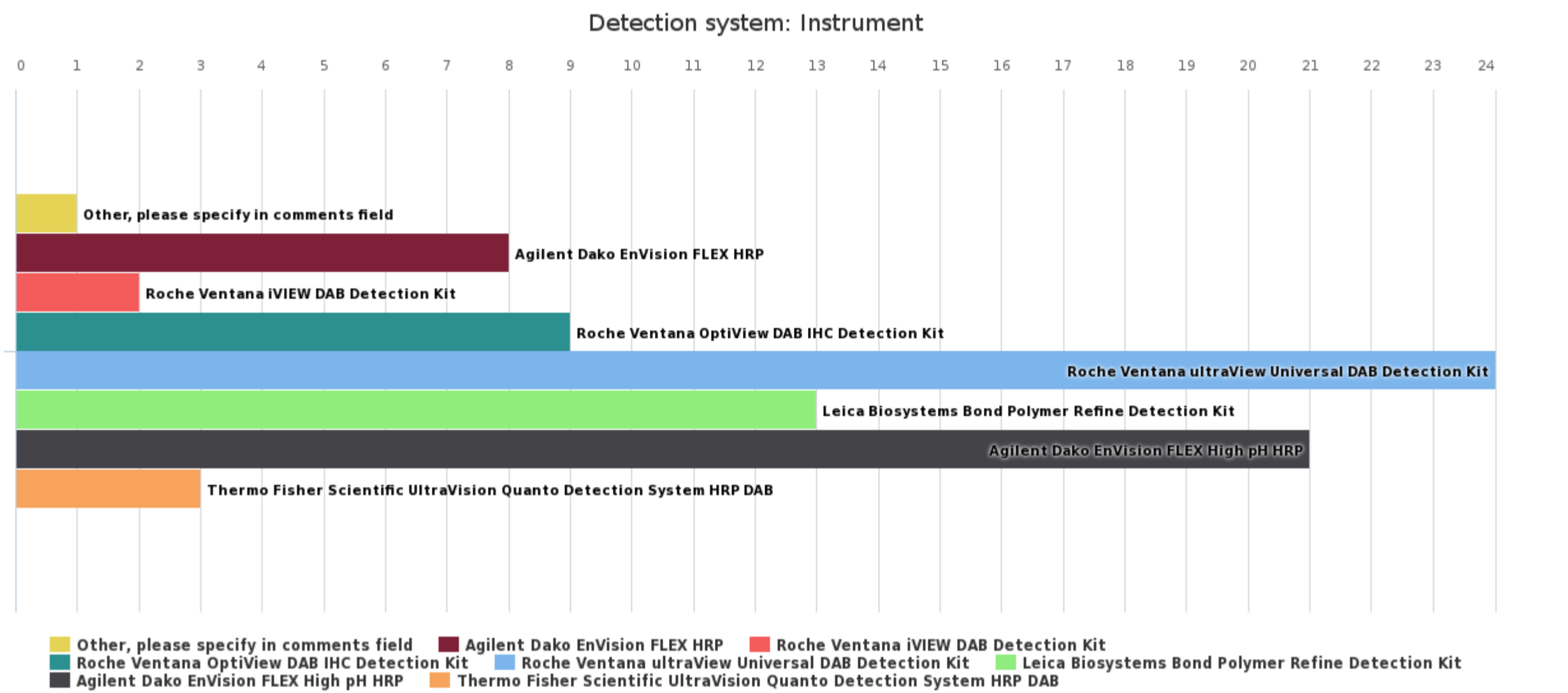
PRETREATMENT

Pretreatment	Pretreatment count
HIER in a microwave oven	3
HIER in a Pt-module	20
<input checked="" type="radio"/> HIER in an instrument	55
Other	3
Total:	81

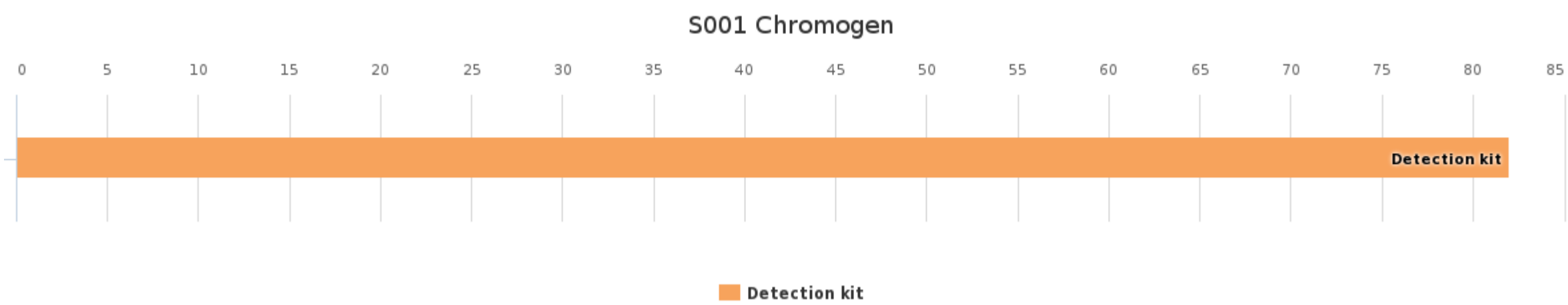


DETECTION SYSTEM

Detection system	Detection system count
Biotin-based	2
Multimer	28
<input checked="" type="radio"/> Polymer	51
Total:	81

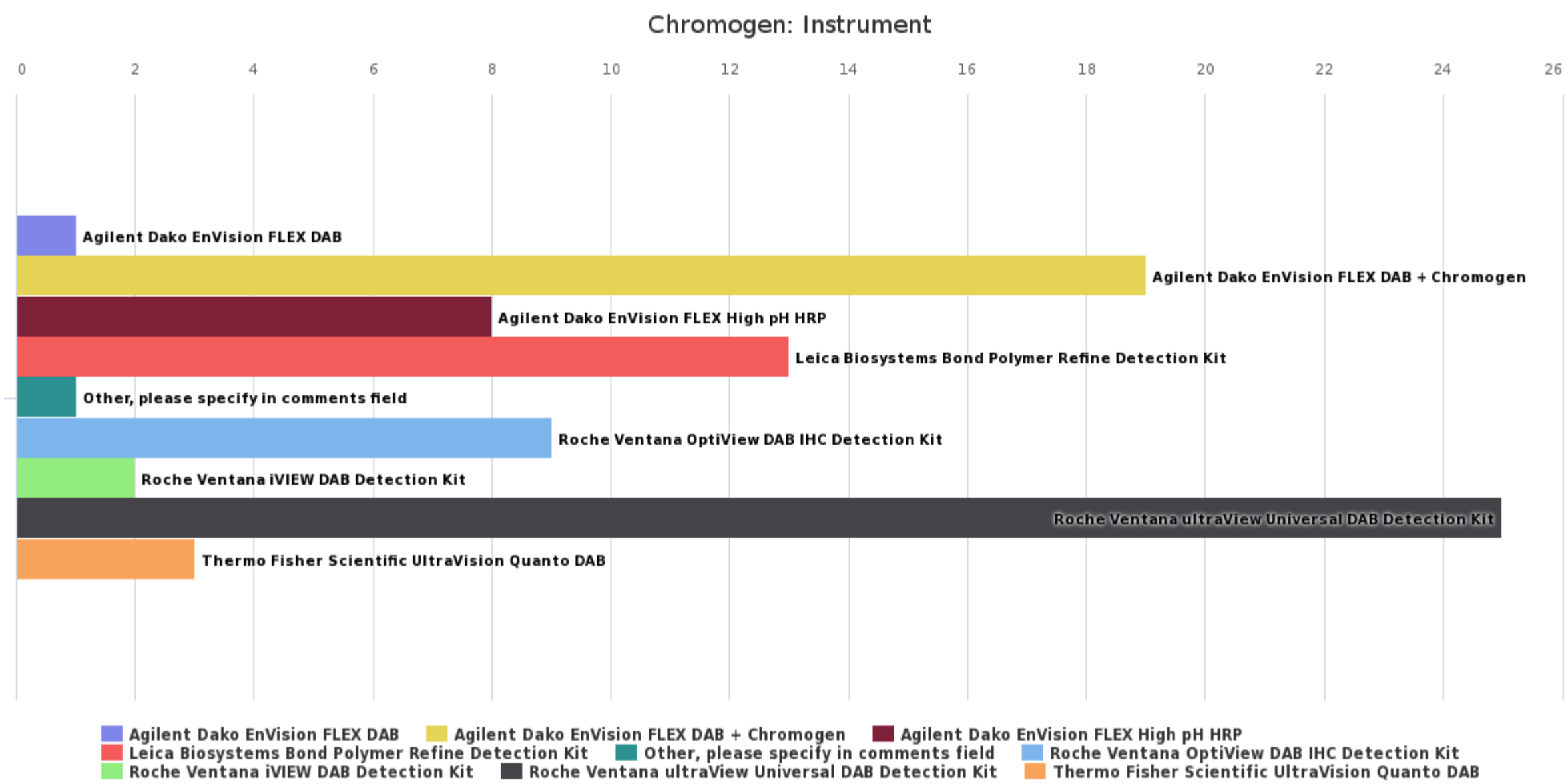


Detection system: Instrument	Detection system: Instrument count
Agilent Dako EnVision FLEX HRP	8
Agilent Dako EnVision FLEX High pH HRP	21
Leica Biosystems Bond Polymer Refine Detection Kit	13
Other, please specify in comments field	1
Roche Ventana OptiView DAB IHC Detection Kit	9
Roche Ventana iVIEW DAB Detection Kit	2
<input checked="" type="radio"/> Roche Ventana ultraView Universal DAB Detection Kit	24
Thermo Fisher Scientific UltraVision Quanto Detection System HRP DAB	3
Total:	81

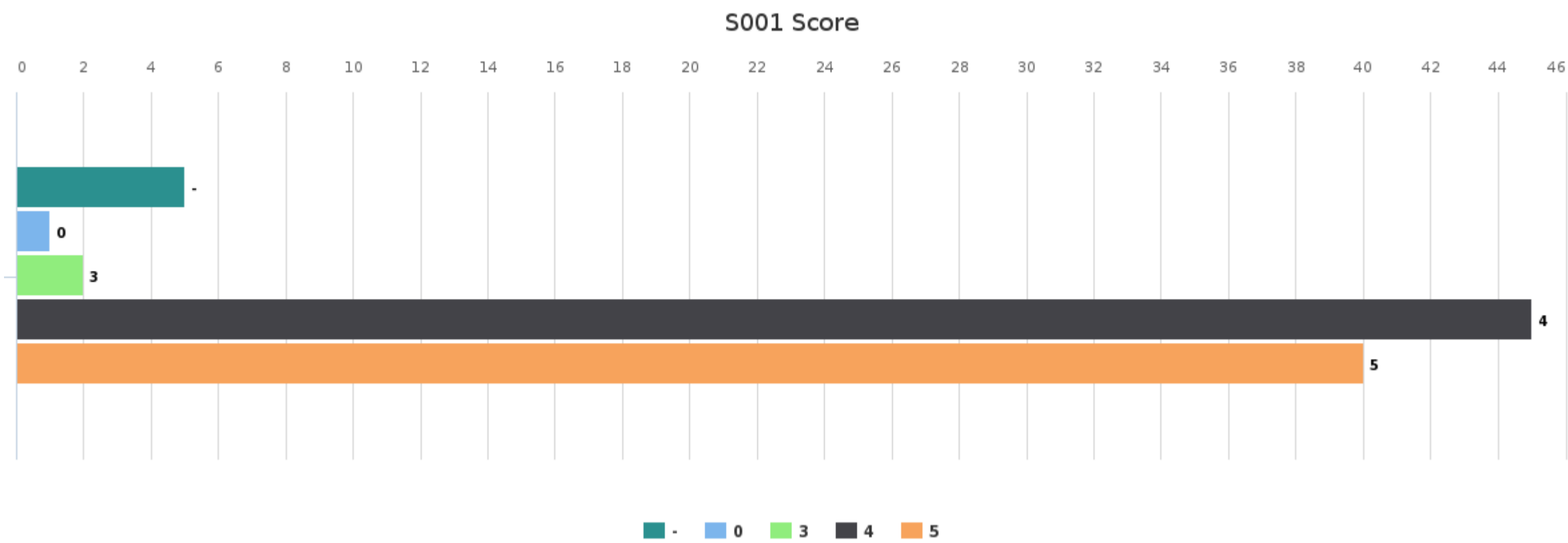


CHROMOGEN

Chromogen	Chromogen count
<input checked="" type="radio"/> Detection kit	82
Total:	82

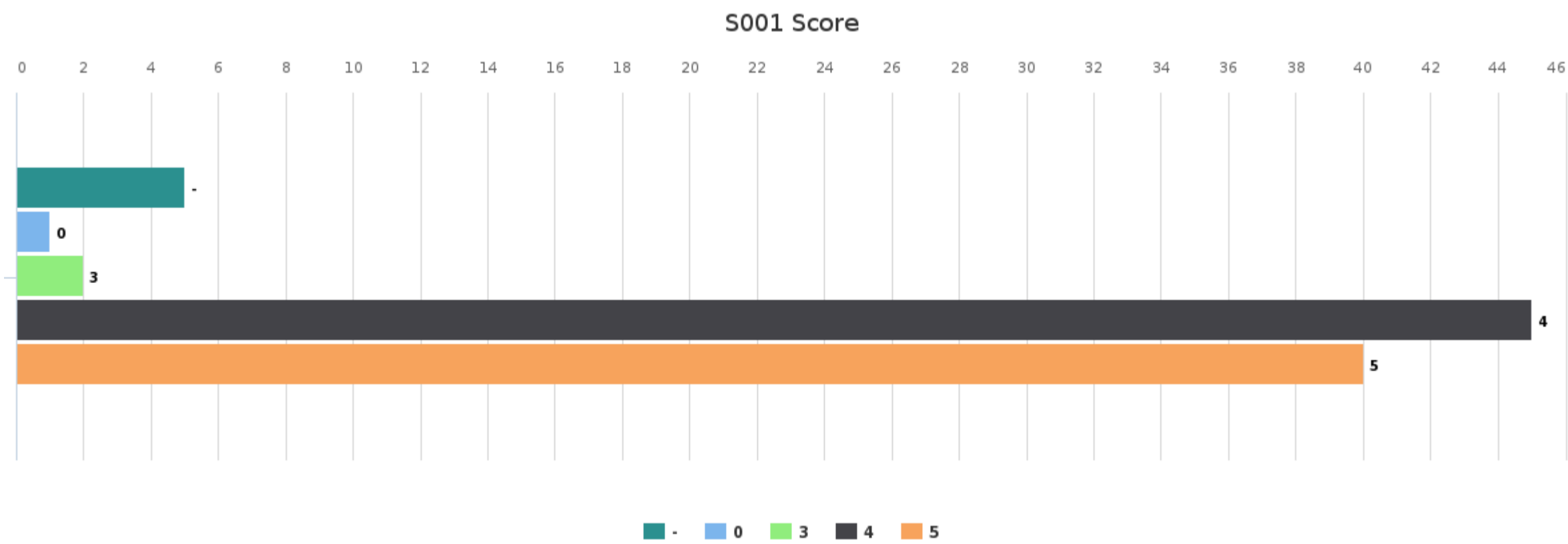


Chromogen: Instrument	Chromogen: Instrument count
Agilent Dako EnVision FLEX DAB	1
Agilent Dako EnVision FLEX DAB + Chromogen	19
Agilent Dako EnVision FLEX High pH HRP	8
Leica Biosystems Bond Polymer Refine Detection Kit	13
Other, please specify in comments field	1
Roche Ventana OptiView DAB IHC Detection Kit	9
Roche Ventana iVIEW DAB Detection Kit	2
<input checked="" type="radio"/> Roche Ventana ultraView Universal DAB Detection Kit	25
Thermo Fisher Scientific UltraVision Quanto DAB	3
Total:	81



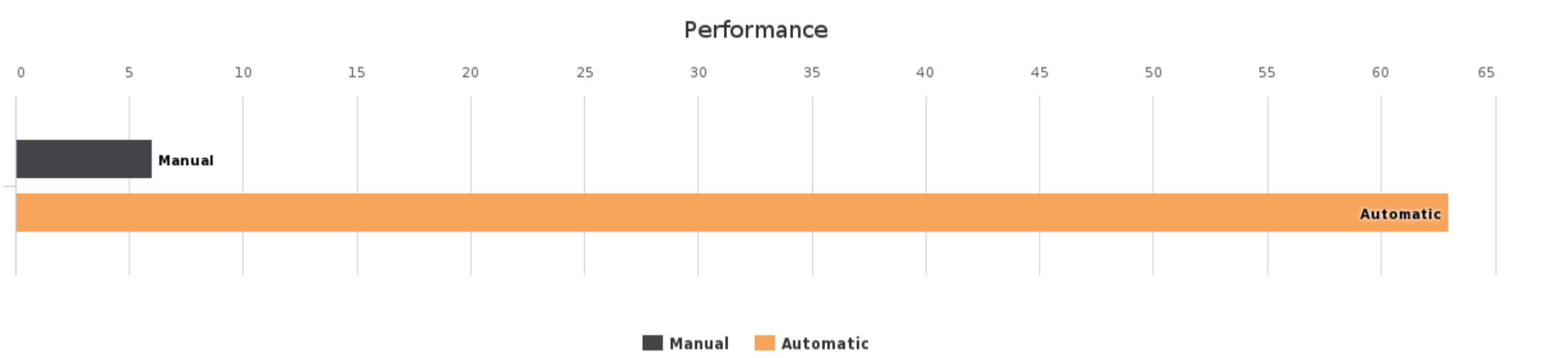
SCORE

Score	Comment
-	
	not available
0	
	negative
3	
	one core is not stained, technical issue?
	uneven, some background staining, one core is missing
4	
	dark hematoxylin
	slightly understained
	slightly understained, uneven
	slightly uneven
	some background staining
	some background staining ,slighly uneven
	some background staining, dark hematoxylin
	some background staining, hematoxylin is quite weak
	some background staining, slightly overstained
	some background staining, slightly uneven
	some background, hematoxylin in pancreas sample is uneven
	some background staining
<input checked="" type="radio"/> 5	
	dark hematoxylin
	<input checked="" type="radio"/> Excellent
Total:	



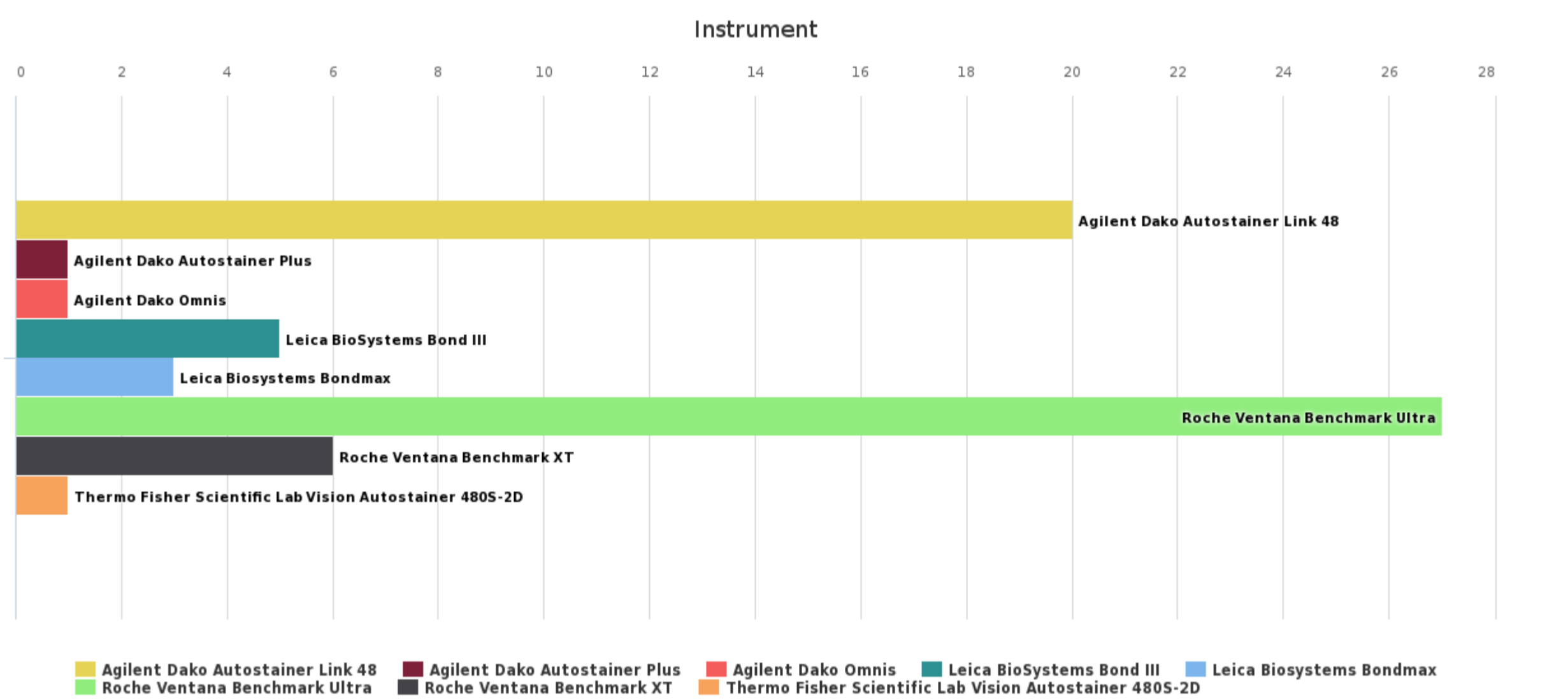
Score	Score count
-	5
0	1
3	2
4	45
<input checked="" type="radio"/> 5	40
Total:	93

S002 | Sample S002 WT1

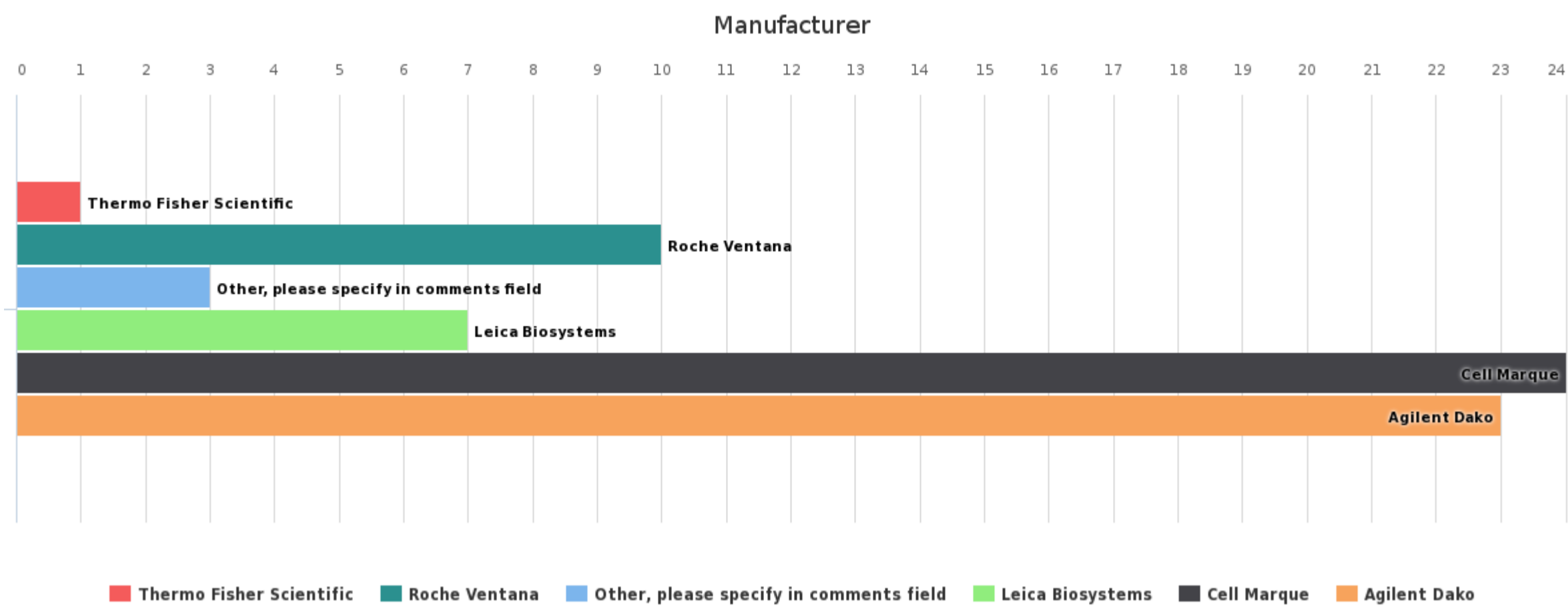


GENERAL DETAILS OF STAINING

Performance	Performance count
<input checked="" type="radio"/> Automatic	63
Manual	6
Total:	69

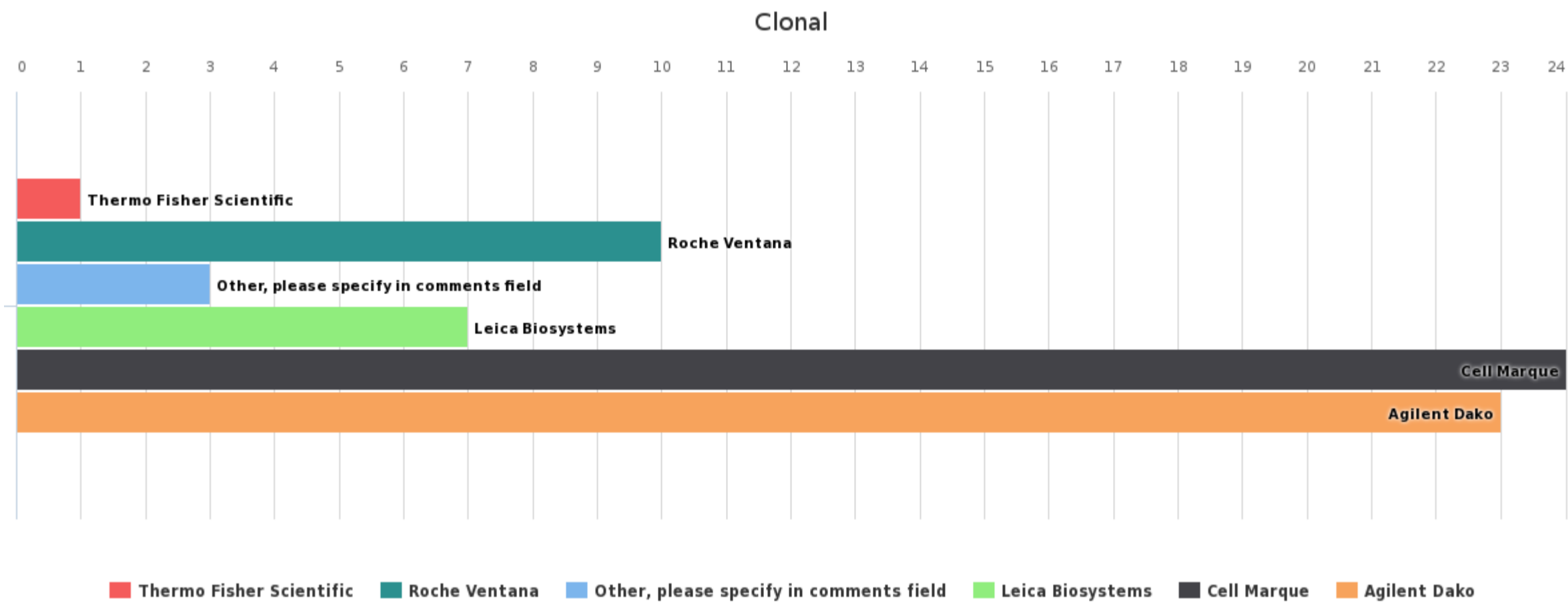


Instrument	Instrument count
Agilent Dako Autostainer Link 48	20
Agilent Dako Autostainer Plus	1
Agilent Dako Omnis	1
Leica BioSystems Bond III	5
Leica Biosystems Bondmax	3
<input checked="" type="radio"/> Roche Ventana Benchmark Ultra	27
Roche Ventana Benchmark XT	6
Thermo Fisher Scientific Lab Vision Autostainer 480S-2D	1
Total:	64

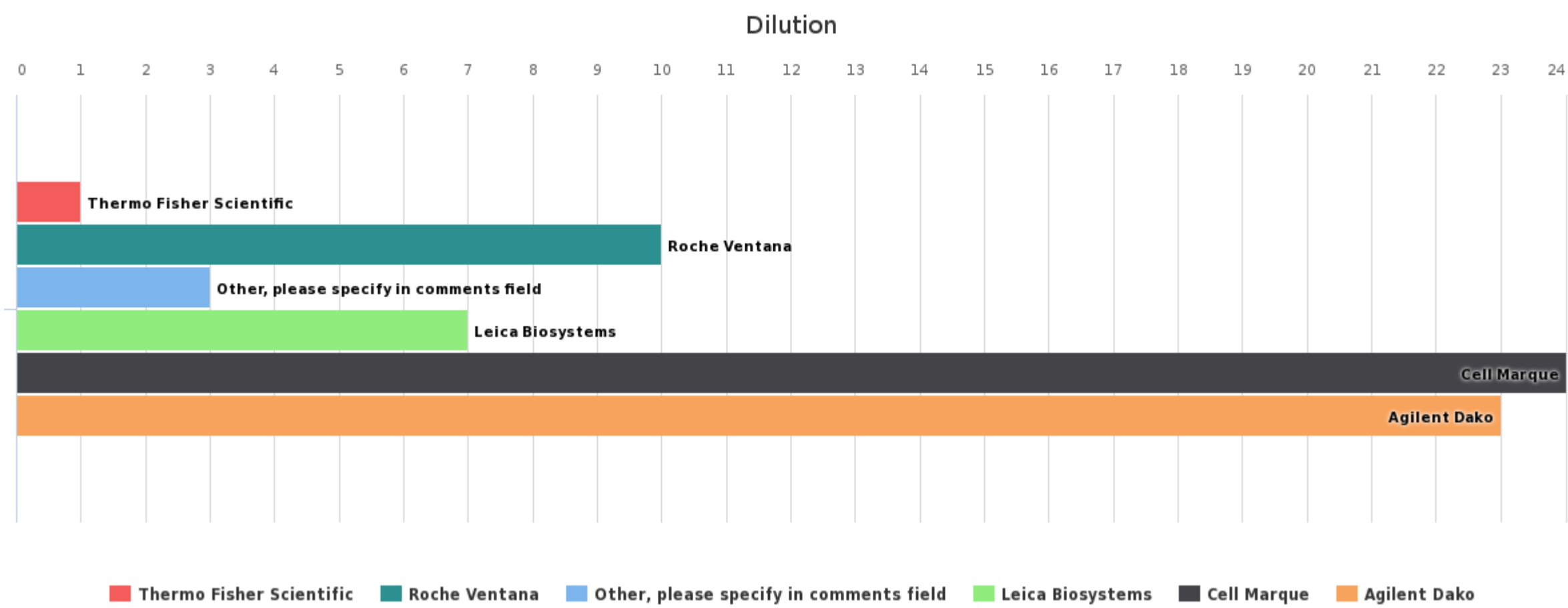


PRIMARY ANTIBODY

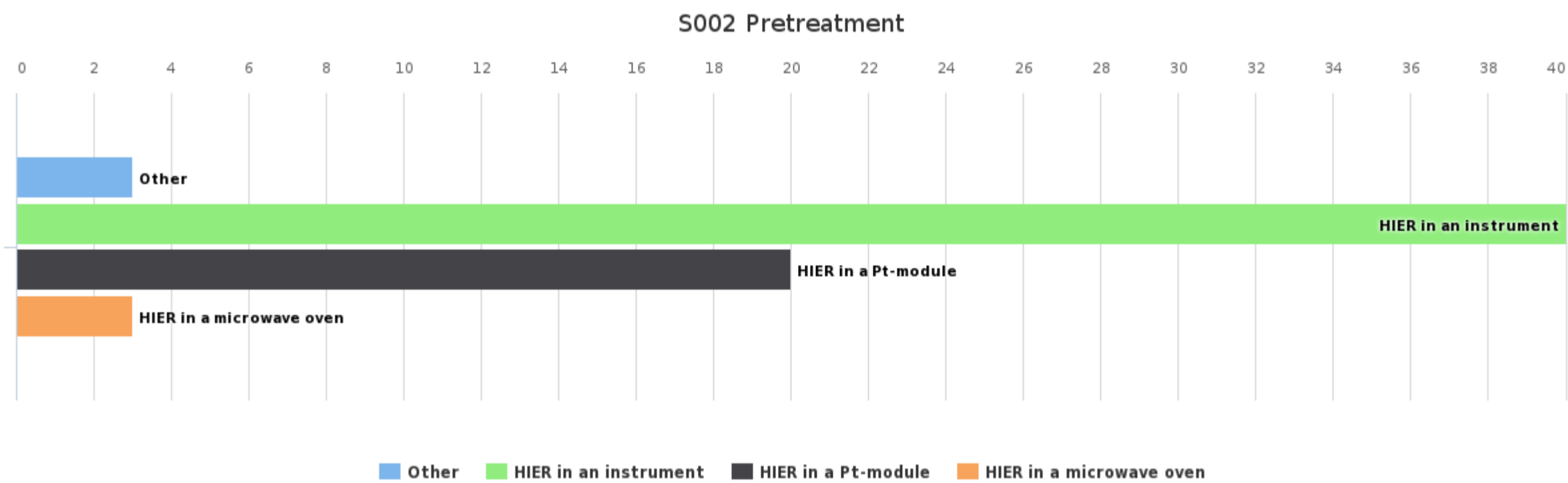
Manufacturer	Manufacturer count
Agilent Dako	23
<input checked="" type="radio"/> Cell Marque	24
Leica Biosystems	7
Other, please specify in comments field	3
Roche Ventana	10
Thermo Fisher Scientific	1
Total:	68



Clonal	Clonal count
<input checked="" type="radio"/> Monoclonal	68
Total:	68

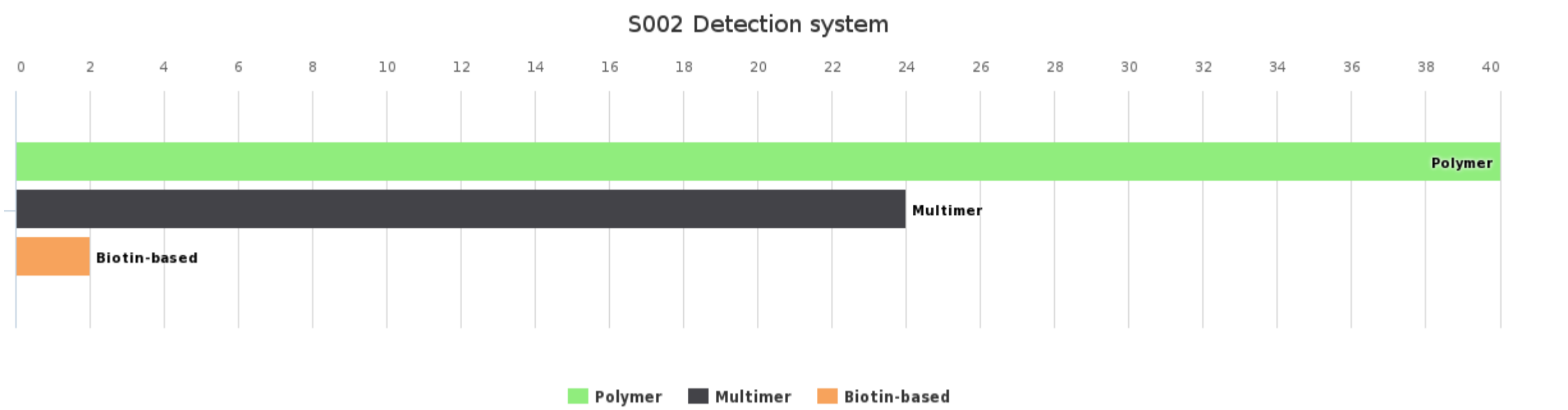


Dilution	Dilution count
Dilution	23
<input checked="" type="radio"/> Ready-to-use	45
Total:	68



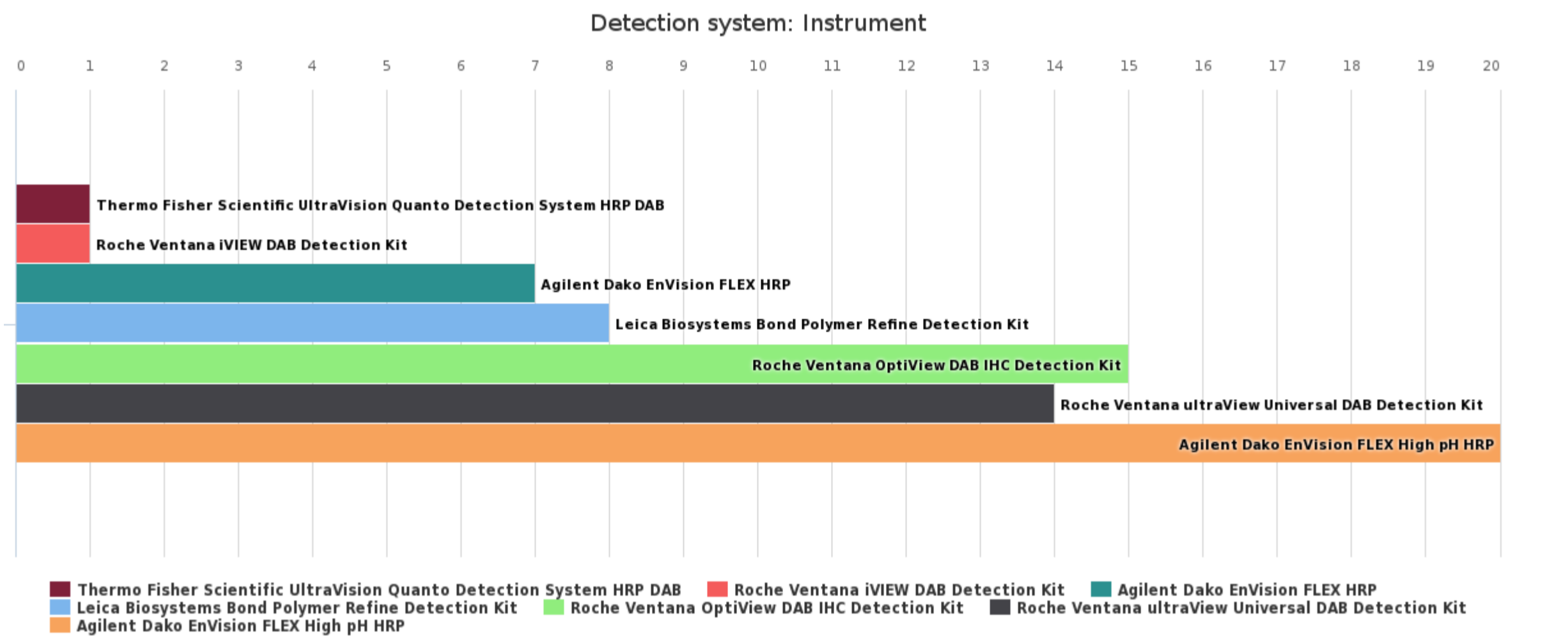
PRETREATMENT

Pretreatment	Pretreatment count
HIER in a microwave oven	3
HIER in a Pt-module	20
<input checked="" type="radio"/> HIER in an instrument	40
Other	3
Total:	66

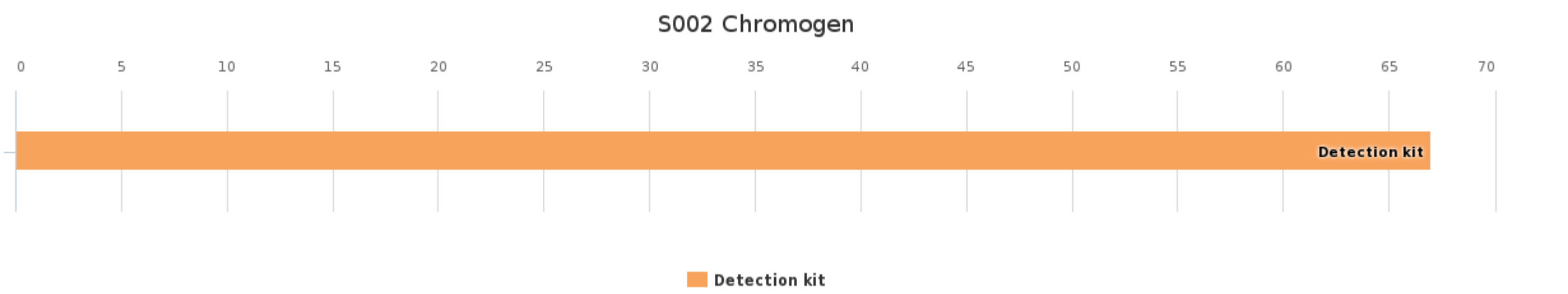


DETECTION SYSTEM

Detection system	Detection system count
Biotin-based	2
Multimer	24
<input checked="" type="radio"/> Polymer	40
Total:	66

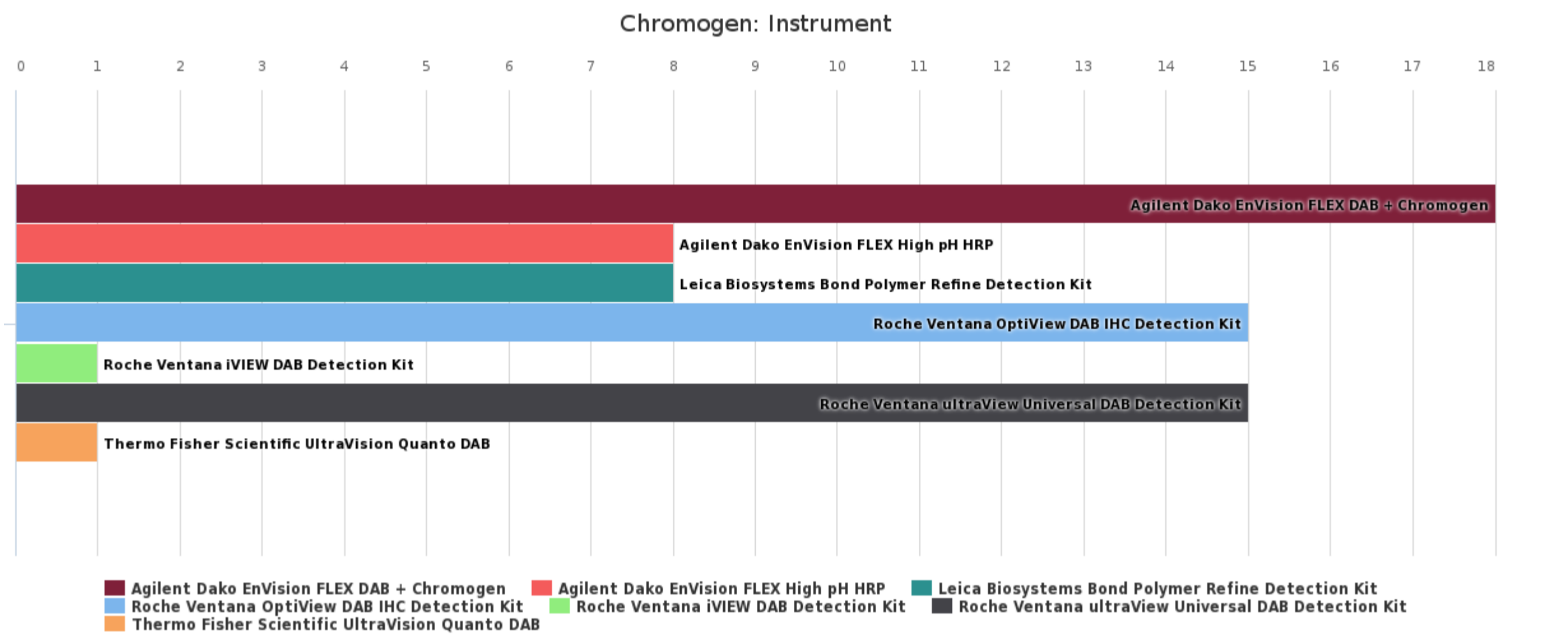


Detection system: Instrument	Detection system: Instrument count
Agilent Dako EnVision FLEX HRP	7
Agilent Dako EnVision FLEX High pH HRP	20
Leica Biosystems Bond Polymer Refine Detection Kit	8
Roche Ventana OptiView DAB IHC Detection Kit	15
Roche Ventana iVIEW DAB Detection Kit	1
<input checked="" type="radio"/> Roche Ventana ultraView Universal DAB Detection Kit	14
Thermo Fisher Scientific UltraVision Quanto Detection System HRP DAB	1
Total:	66

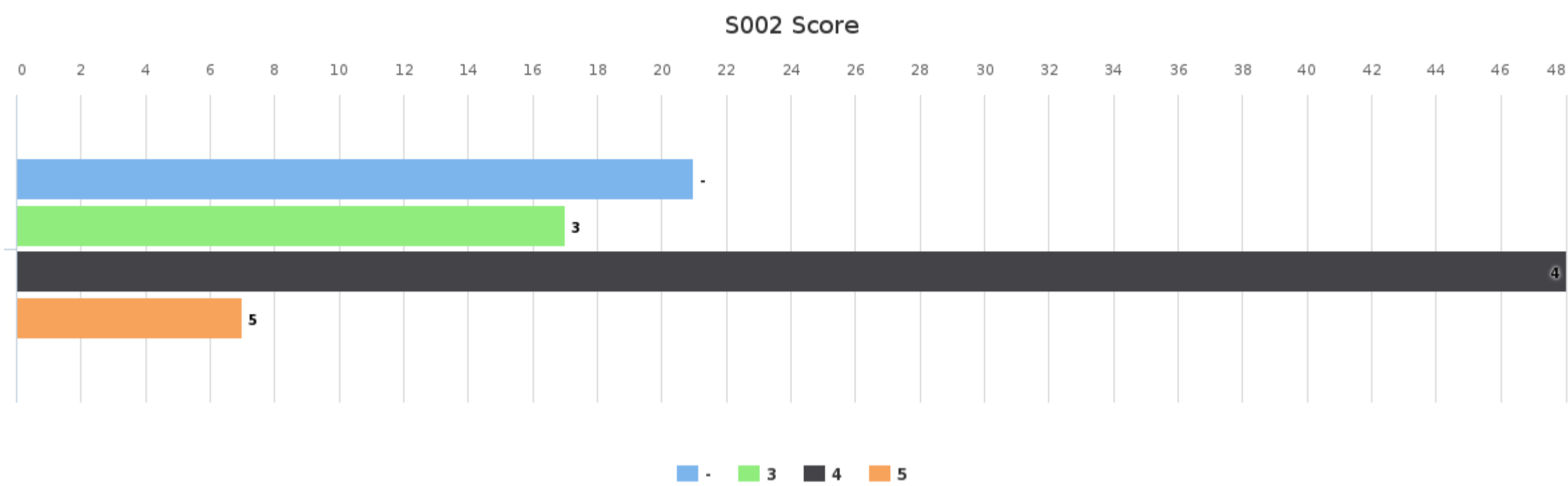


CHROMOGEN

Chromogen	Chromogen count
<input checked="" type="radio"/> Detection kit	67
Total:	67

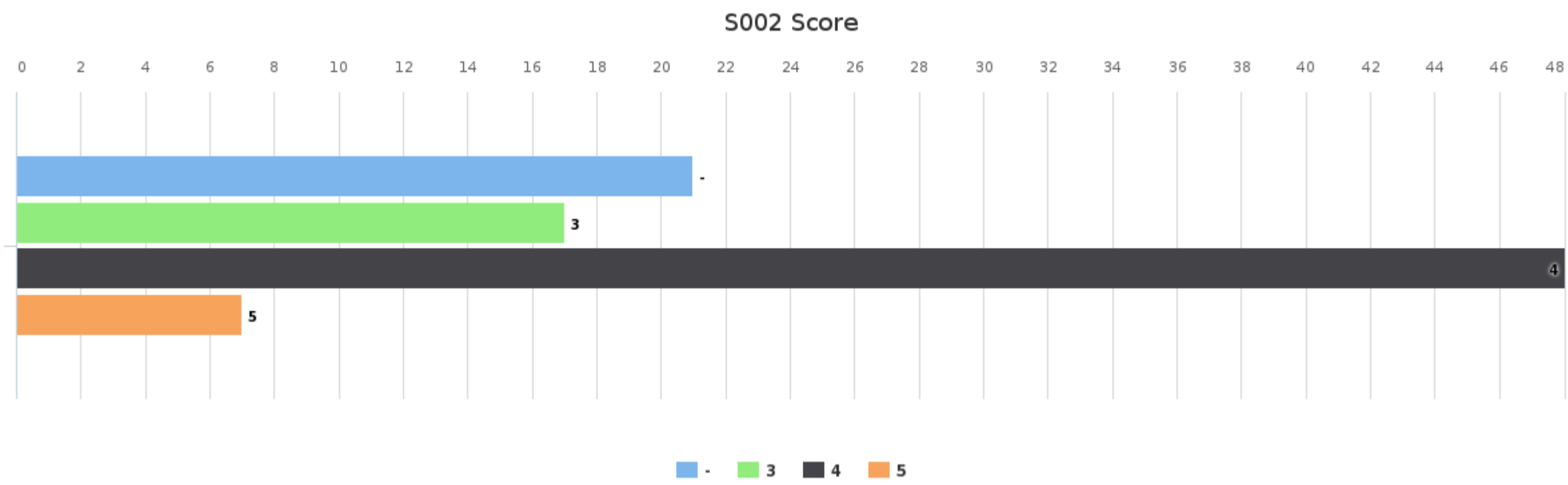


Chromogen: Instrument	Chromogen: Instrument count
Agilent Dako EnVision FLEX DAB + Chromogen	18
Agilent Dako EnVision FLEX High pH HRP	8
Leica Biosystems Bond Polymer Refine Detection Kit	8
Roche Ventana OptiView DAB IHC Detection Kit	15
Roche Ventana iVIEW DAB Detection Kit	1
<input checked="" type="radio"/> Roche Ventana ultraView Universal DAB Detection Kit	15
Thermo Fisher Scientific UltraVision Quanto DAB	1
Total:	66



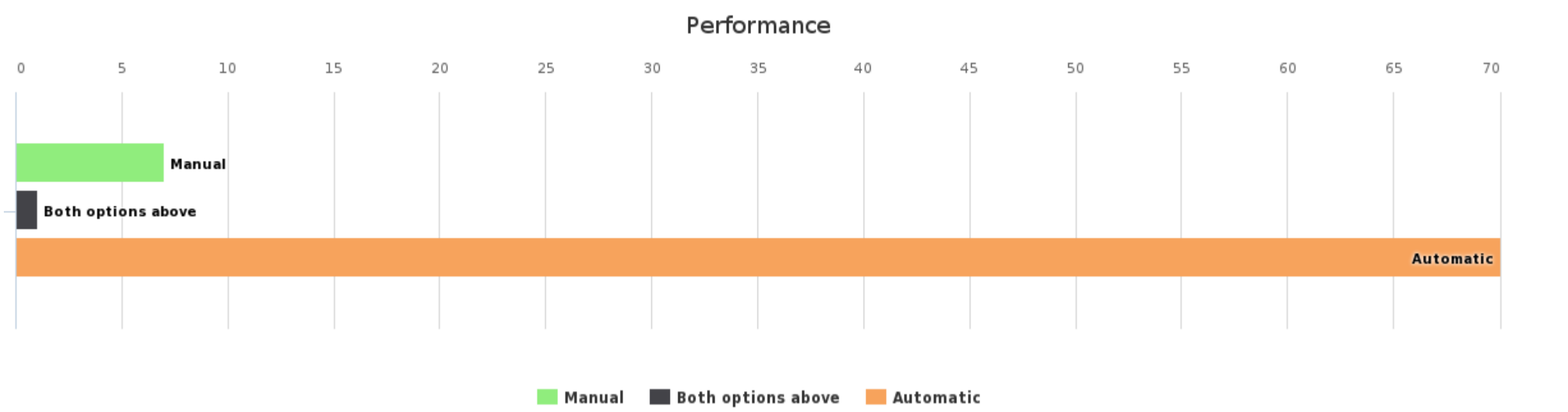
SCORE

Score	Comment
-	
	not available
3	
	background staining in kidney sample
	background staining in kidney sample, overstained
	contaminated from some other sample?, some background staining, stain deposits
	overstained
	some background staining in kidney sample, overstained
	some background staining in kidney sample, slightly overstained
	some background staining in kidney sample, understained, dark hematoxylin
	some background staining in kidney sample, uneven
	some background staining, uneven
	understained
	understained, dark hematoxylin, some background staining in kidney sample
	understained, false positive staining in fallobian tube
	understained, uneven
	undertaining
<input checked="" type="radio"/> 4	
	background staining in kidney sample
	hematoxylin is quite weak
	slightly overstained, some background staining in kidney sample
	slightly overstained, uneven
	<input checked="" type="radio"/> slightly understained
	slightly understained, some background staining in kidney sample
	slightly understained, uneven
	slightly uneven
	sligtly understained
	some background in kidney sample, slightly understained
	some background in kidney sample, sligthly understained
	some background staining
	some background staining in kidney sample
	some background staining in kidney sample, follobian tube sample is uneven
	some background staining in kidney sample, slightly overstained
	some background staining in kidney sample, slightly understained
	some background staining in kidney sample, uneven staining in epithelial cells in fallobian tube
	some background staining, slightly eneven
	uneven staining in epithelial cells in fallobian tube
5	
	Excellent
Total:	



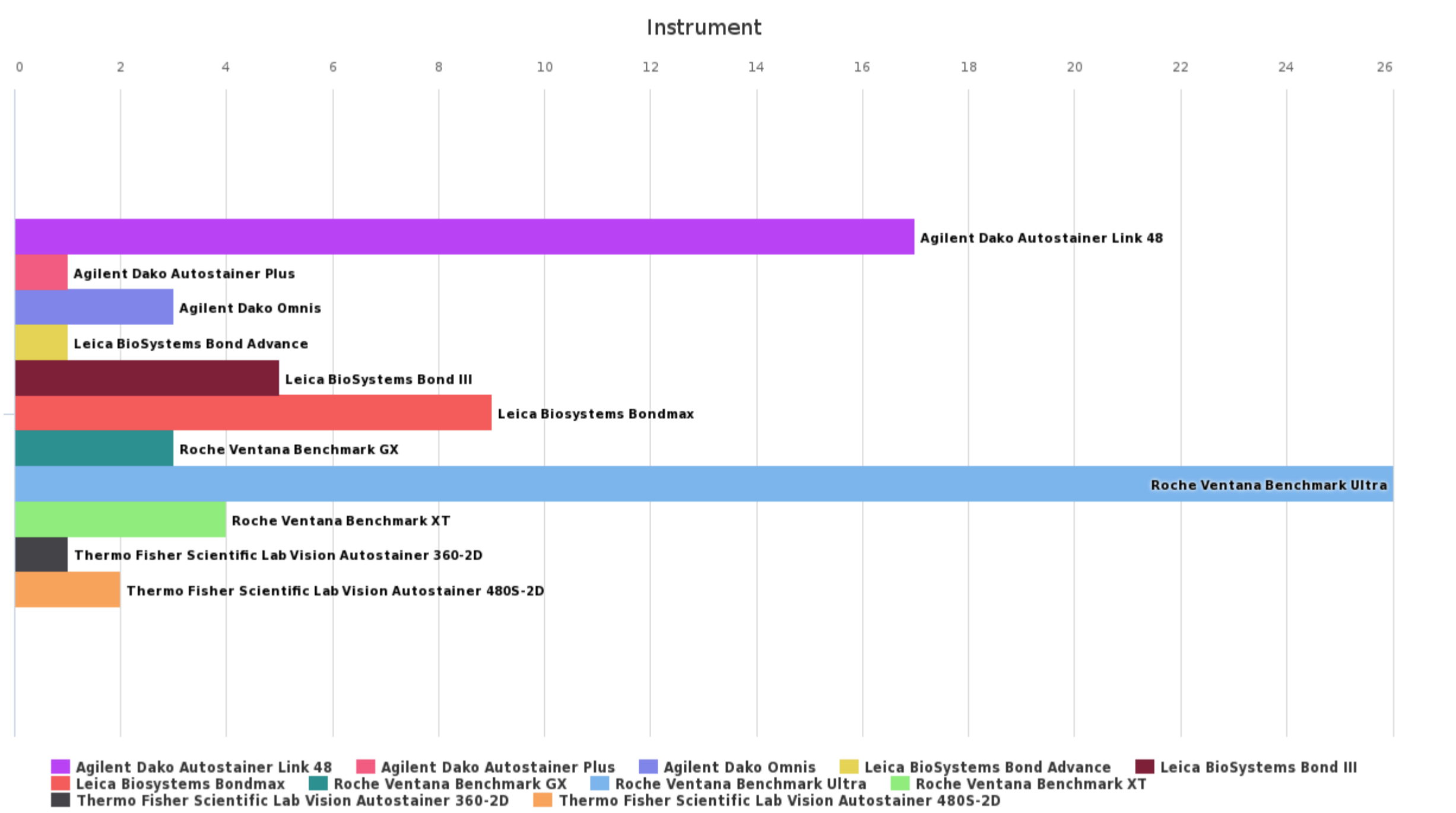
Score	Score count
-	21
3	17
<input checked="" type="radio"/> 4	48
5	7
Total:	93

S003 | Sample S003 CK20



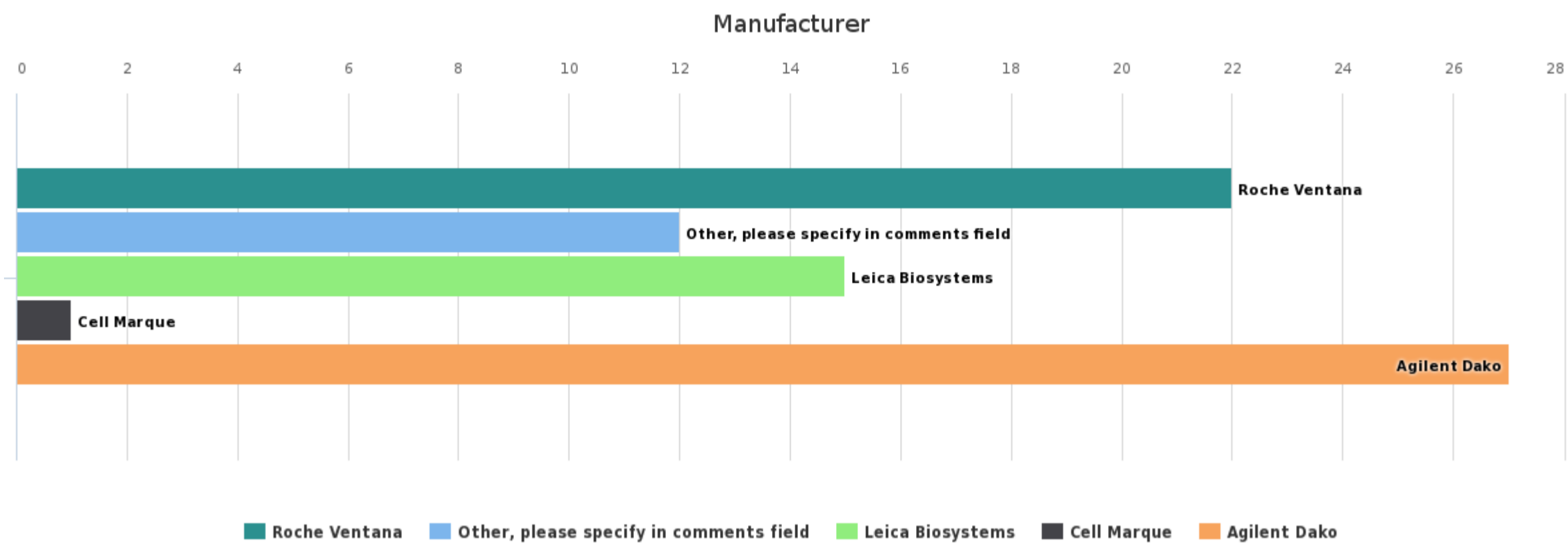
GENERAL DETAILS OF STAINING

Performance	Performance count
<input checked="" type="radio"/> Automatic	70
Both options above	1
Manual	7
Total:	78



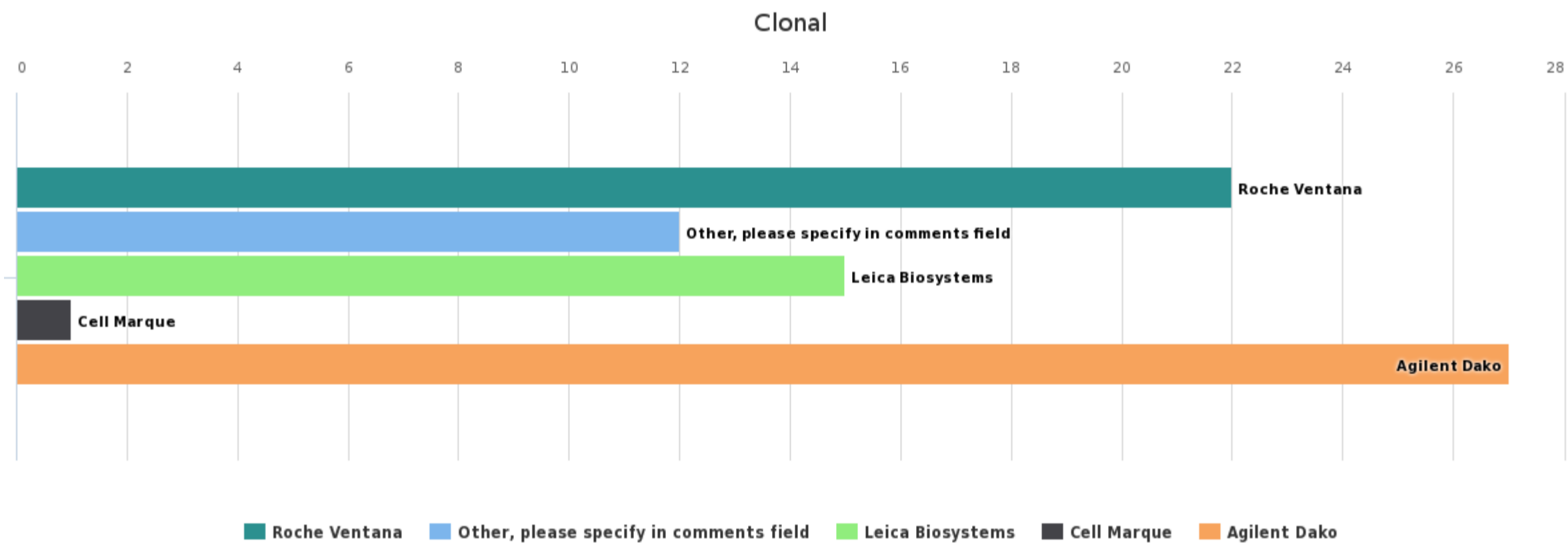
Instrument	Instrument count
Agilent Dako Autostainer Link 48	17
Agilent Dako Autostainer Plus	1
Agilent Dako Omnis	3
Leica BioSystems Bond Advance	1
Leica BioSystems Bond III	5
Leica Biosystems Bondmax	9
Roche Ventana Benchmark GX	3
<input checked="" type="radio"/> Roche Ventana Benchmark Ultra	26

Roche Ventana Benchmark XT	4
Thermo Fisher Scientific Lab Vision Autostainer 360-2D	1
Thermo Fisher Scientific Lab Vision Autostainer 480S-2D	2
Total:	72

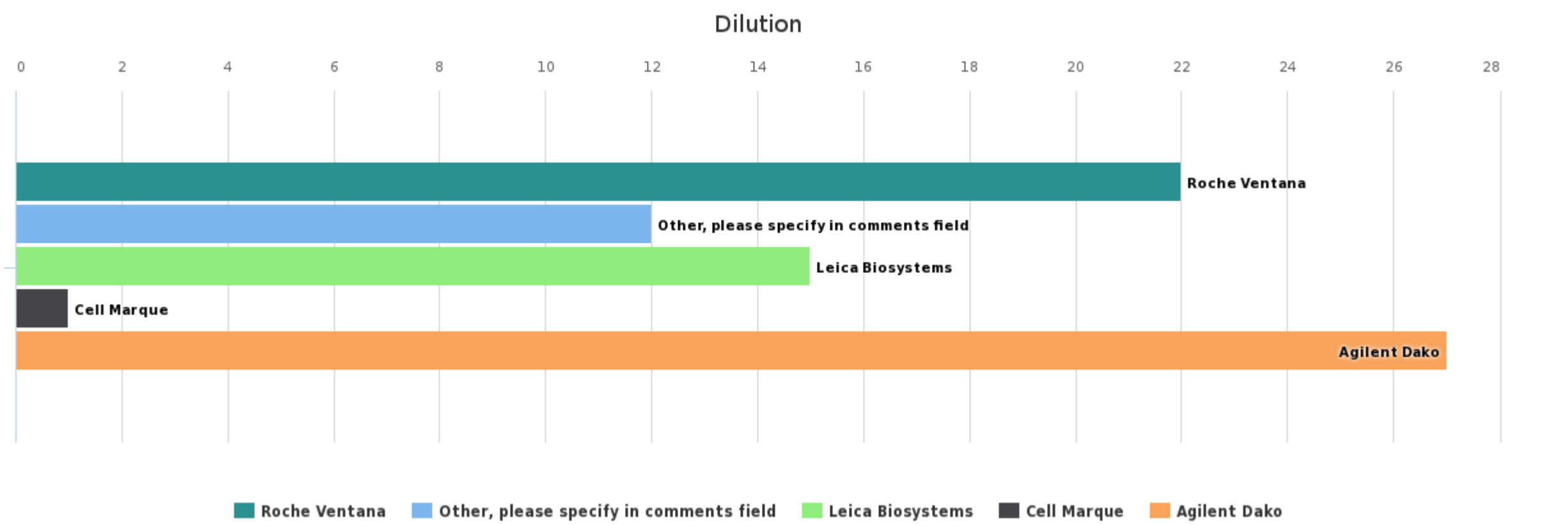


PRIMARY ANTIBODY

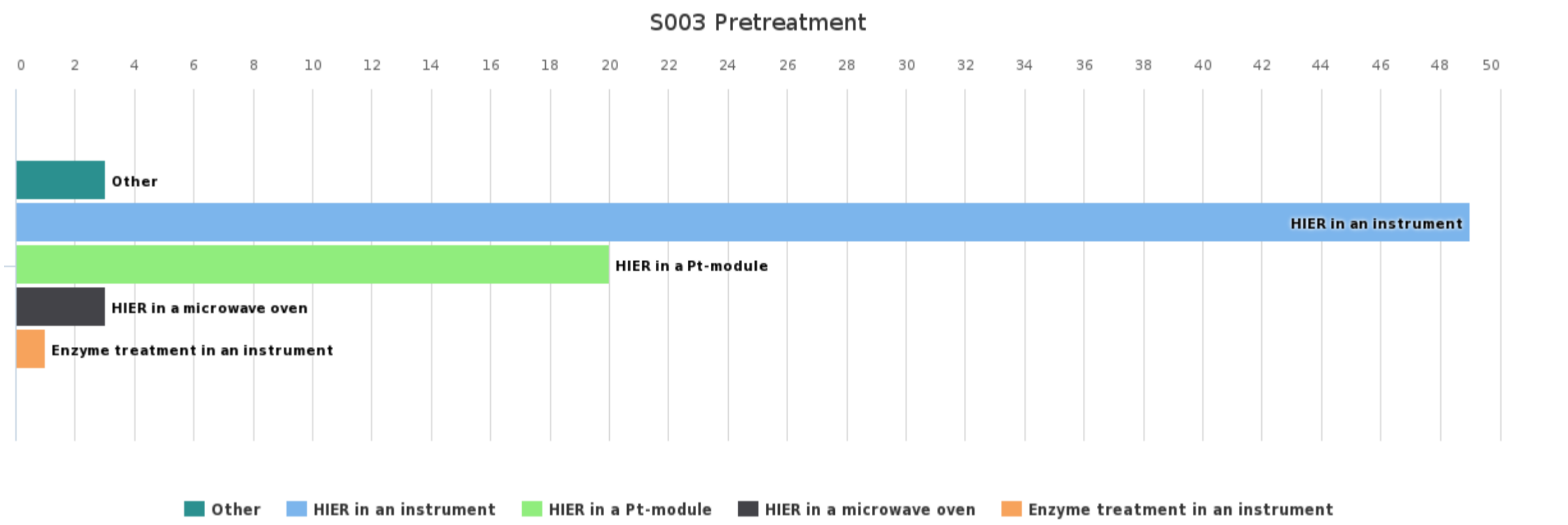
Manufacturer	Manufacturer count
Agilent Dako	27
Cell Marque	1
Leica Biosystems	15
Other, please specify in comments field	12
<input checked="" type="radio"/> Roche Ventana	22
Total:	77



Clonal	Clonal count
<input checked="" type="radio"/> Monoclonal	75
Polyclonal	1
Total:	76

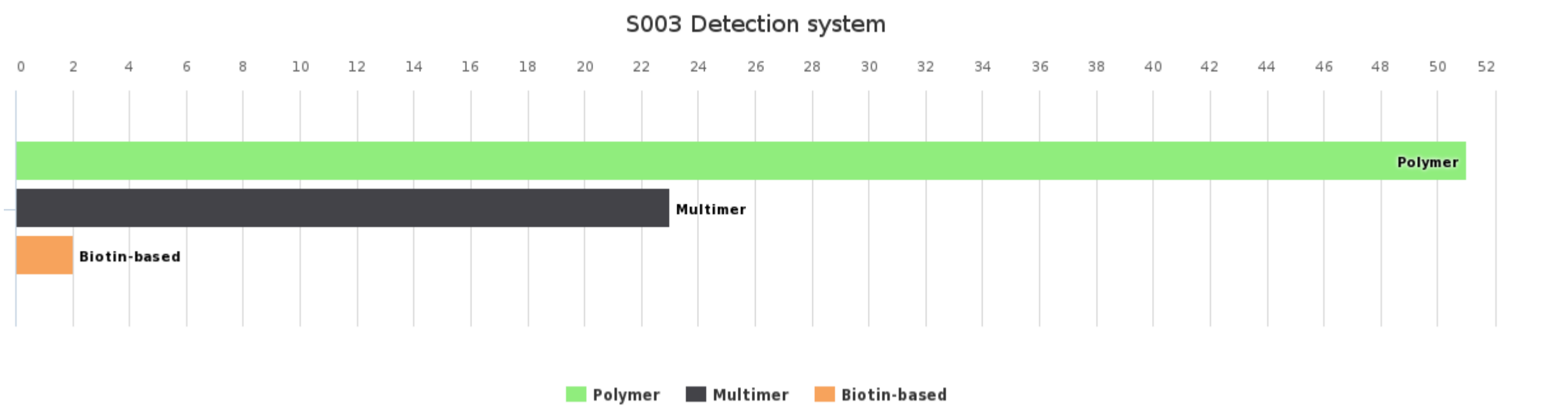


Dilution	Dilution count
Dilution	31
<input checked="" type="radio"/> Ready-to-use	46
Total:	77



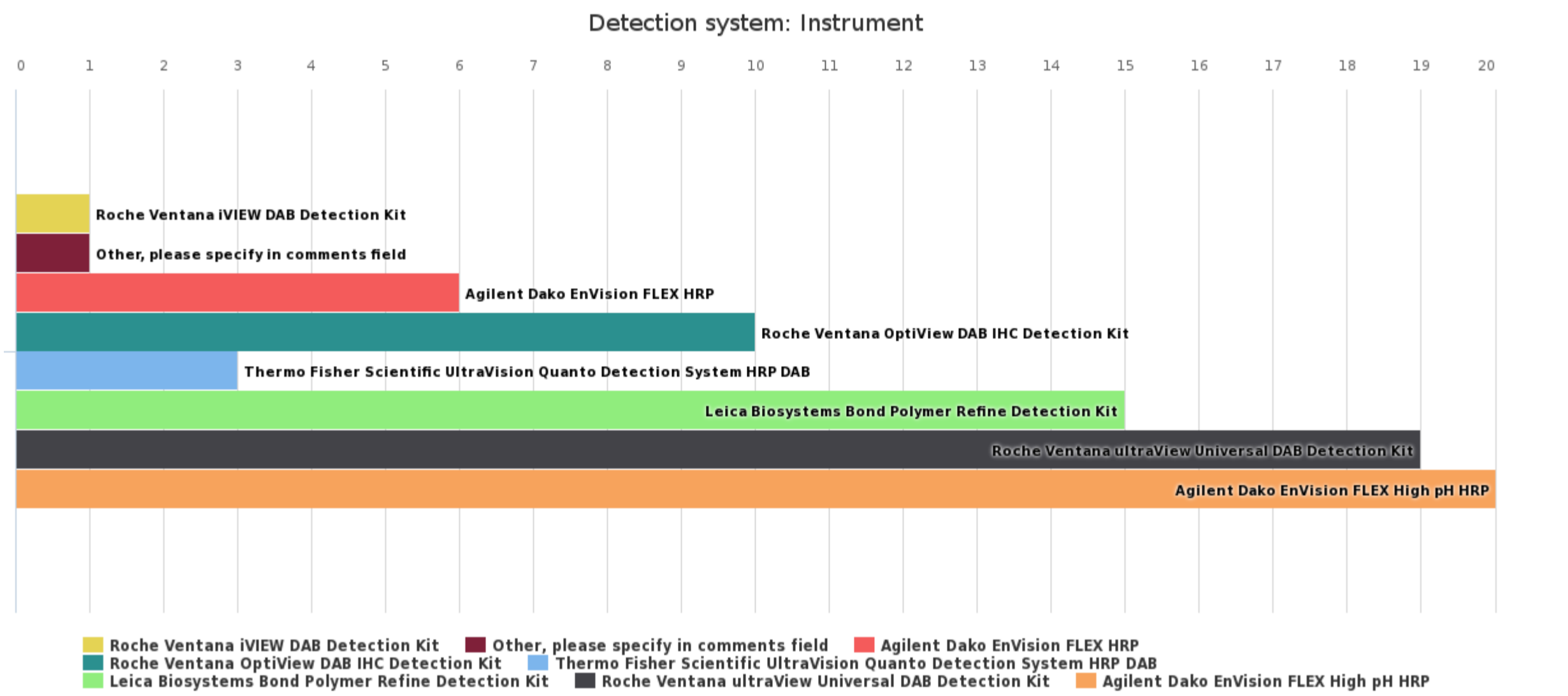
PRETREATMENT

Pretreatment	Pretreatment count
Enzyme treatment in an instrument	1
HIER in a microwave oven	3
HIER in a Pt-module	20
<input checked="" type="radio"/> HIER in an instrument	49
Other	3
Total:	76

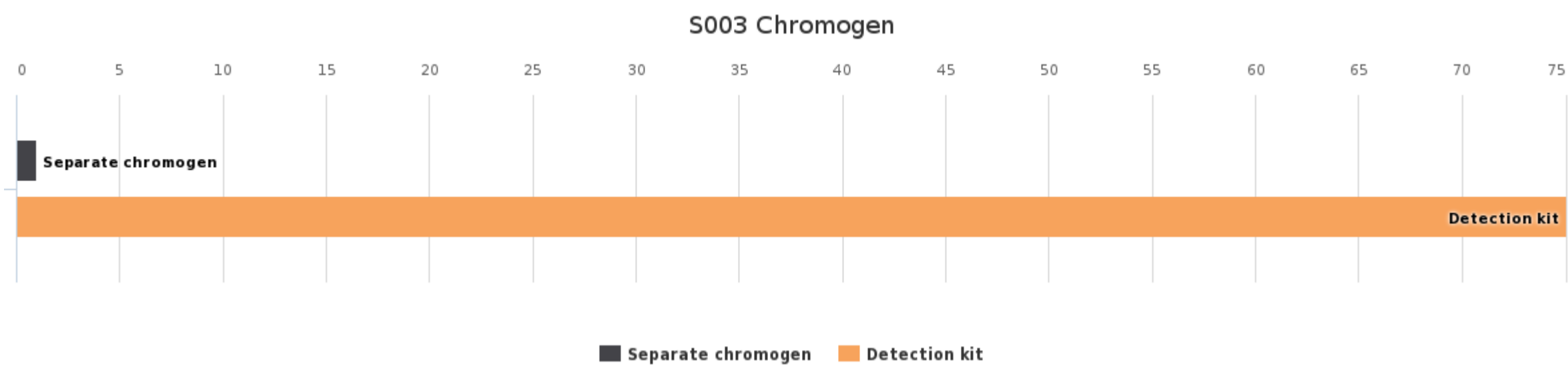


DETECTION SYSTEM

Detection system	Detection system count
Biotin-based	2
Multimer	23
<input checked="" type="radio"/> Polymer	51
Total:	76

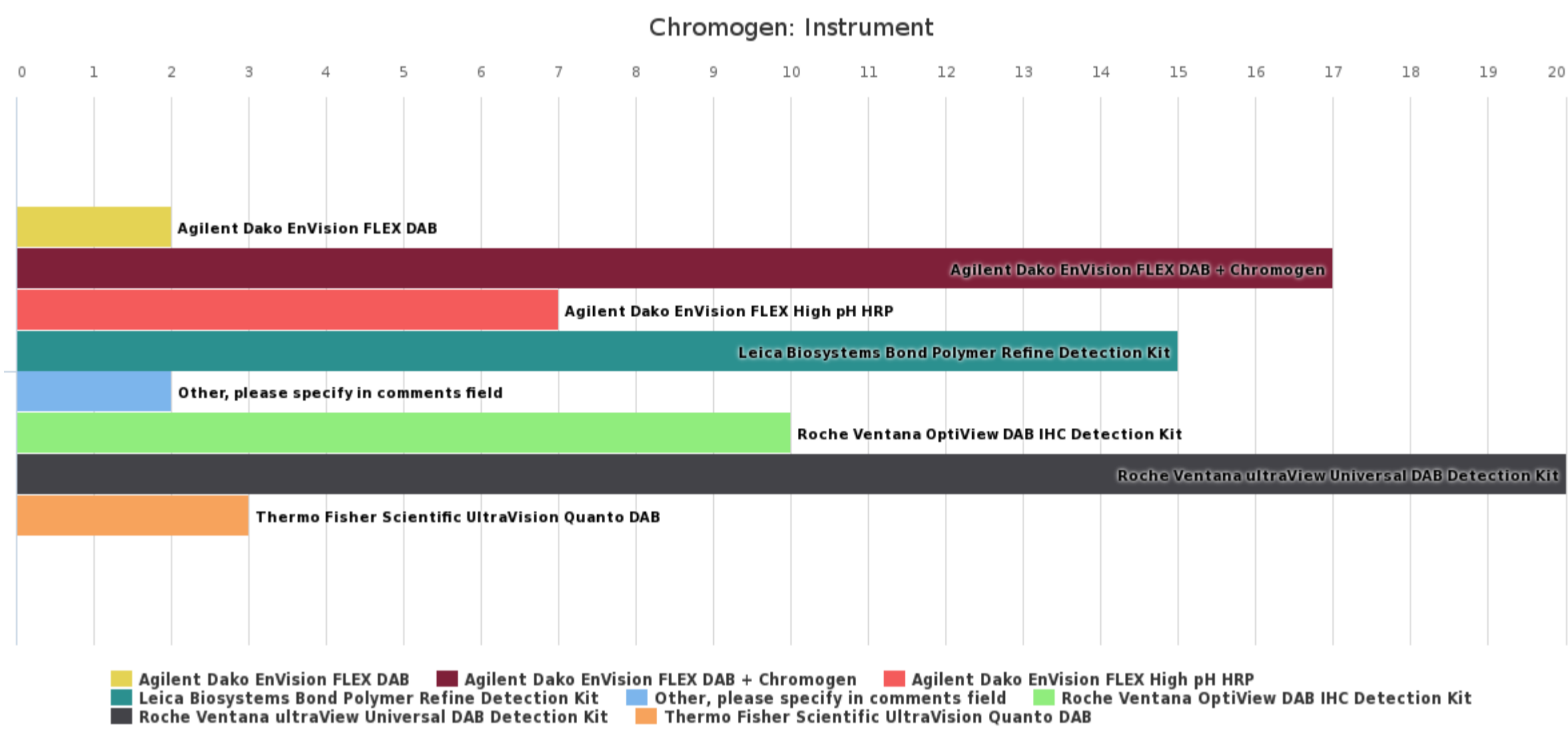


Detection system: Instrument	Detection system: Instrument count
Agilent Dako EnVision FLEX HRP	6
Agilent Dako EnVision FLEX High pH HRP	20
Leica Biosystems Bond Polymer Refine Detection Kit	15
Other, please specify in comments field	1
Roche Ventana OptiView DAB IHC Detection Kit	10
Roche Ventana iVIEW DAB Detection Kit	1
<input checked="" type="radio"/> Roche Ventana ultraView Universal DAB Detection Kit	19
Thermo Fisher Scientific UltraVision Quanto Detection System HRP DAB	3
Total:	75

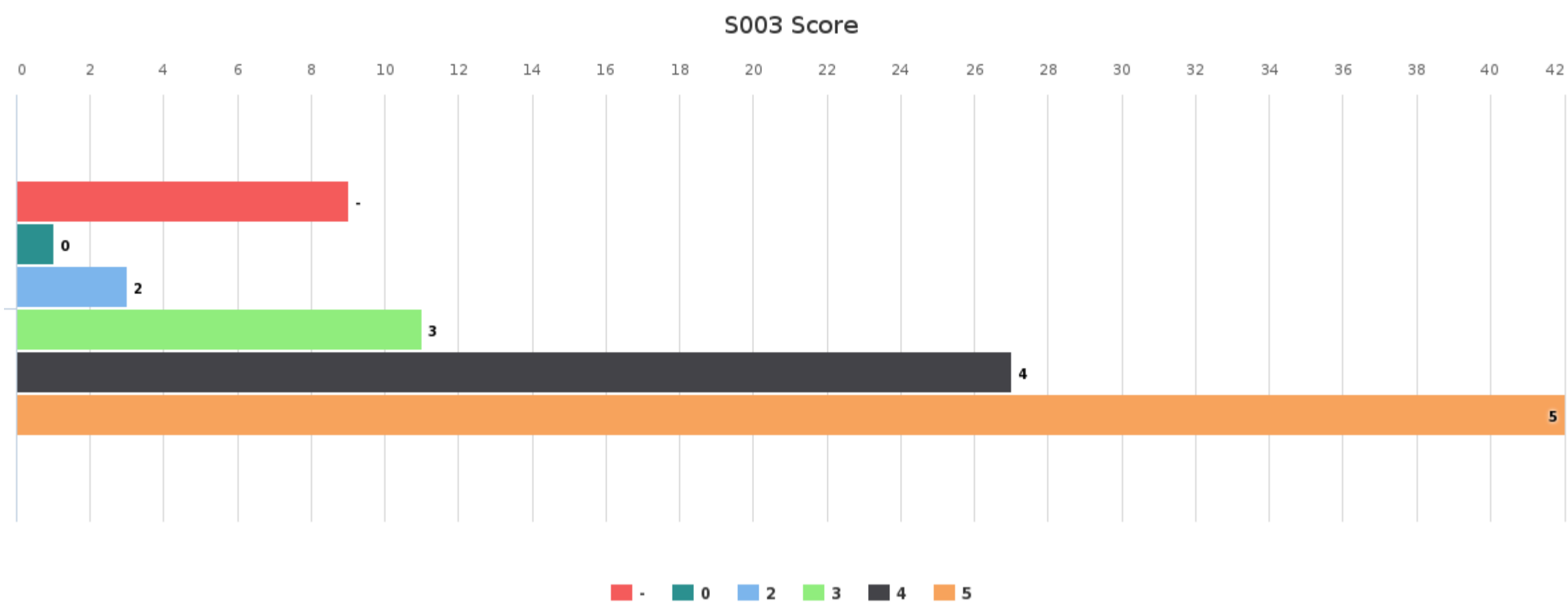


CHROMOGEN

Chromogen	Chromogen count
<input checked="" type="radio"/> Detection kit	75
Separate chromogen	1
Total:	76

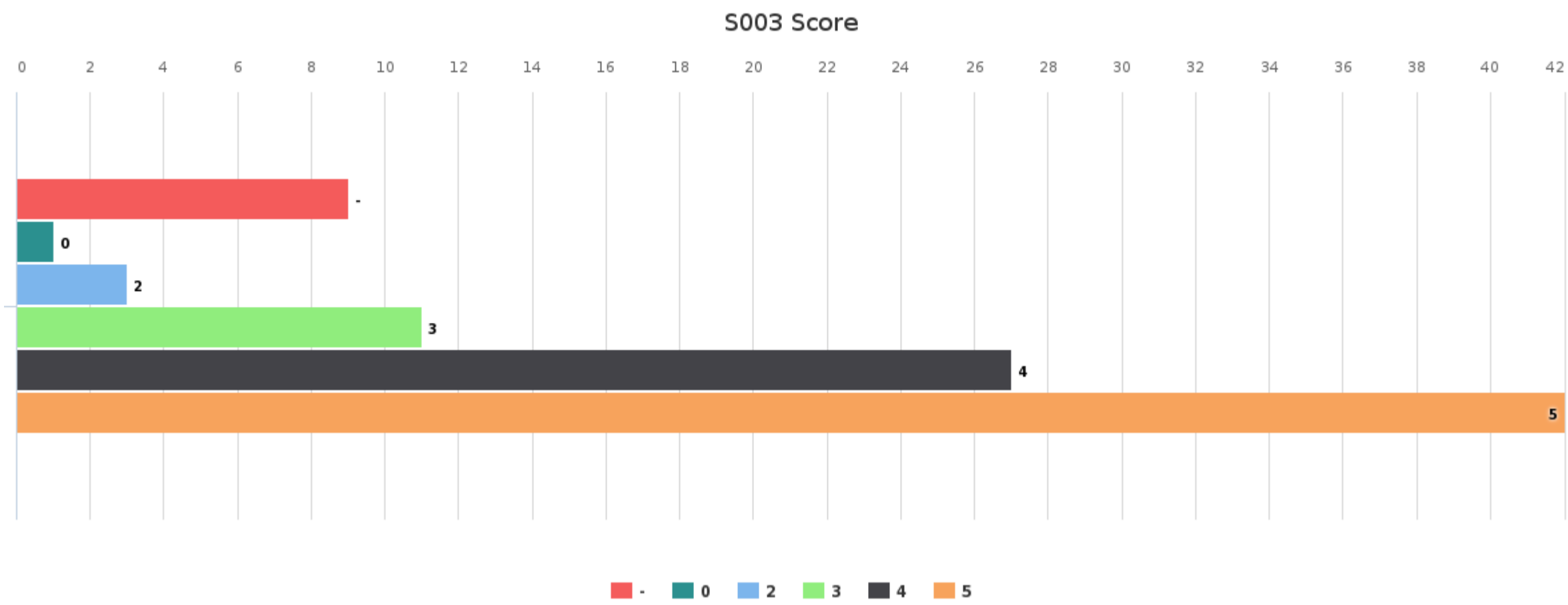


Chromogen: Instrument	Chromogen: Instrument count
Agilent Dako EnVision FLEX DAB	2
Agilent Dako EnVision FLEX DAB + Chromogen	17
Agilent Dako EnVision FLEX High pH HRP	7
Leica Biosystems Bond Polymer Refine Detection Kit	15
Other, please specify in comments field	2
Roche Ventana OptiView DAB IHC Detection Kit	10
<input checked="" type="radio"/> Roche Ventana ultraView Universal DAB Detection Kit	20
Thermo Fisher Scientific UltraVision Quanto DAB	3
Total:	76



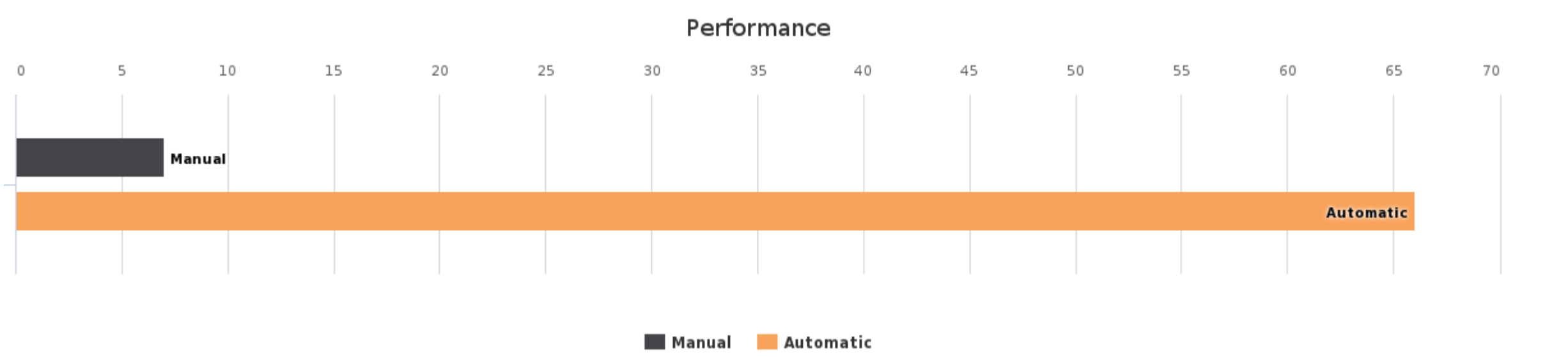
SCORE

Score	Comment
-	
	not available
0	
	negative
2	
	false positivity in muscle cells
	understained, uneven, background staining
	weak staining
3	
	slightly understained, stain deposits
	understained
	understained, dark hematoxylin
	understained, slightly dark hematoxylin
	understaining
	undertained
<input checked="" type="radio"/> 4	
	<input checked="" type="radio"/> slightly understained
	slightly understained, dark hematoxylin
	slightly understained, some background staining
	weak hematoxylin in tumor sample
5	
	Excellent
	hematoxylin is quite weak
Total:	



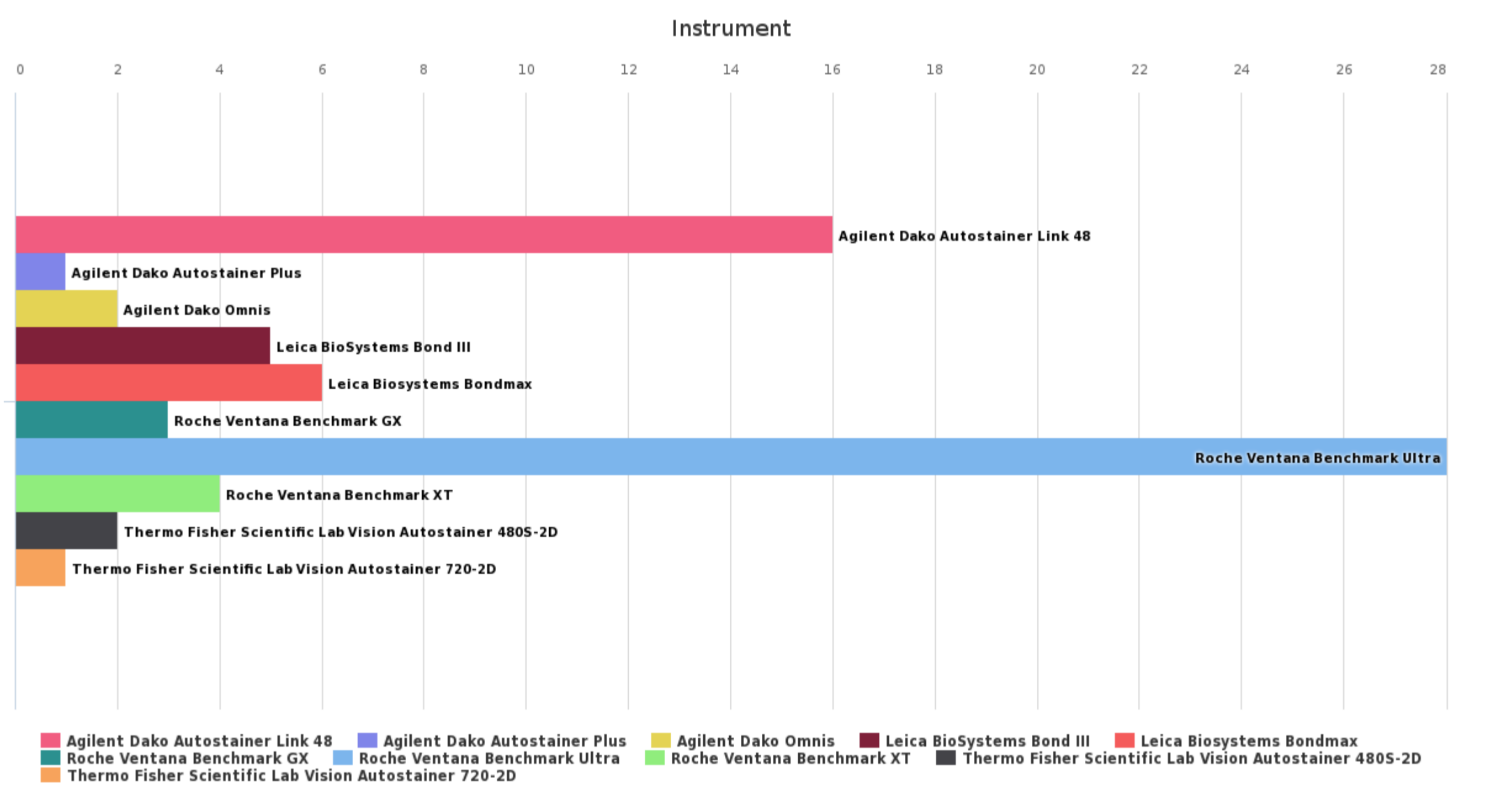
Score	Score count
-	9
0	1
2	3
3	11
<input checked="" type="radio"/> 4	27
5	42
Total:	93

S004 | Sample S004 CDX2

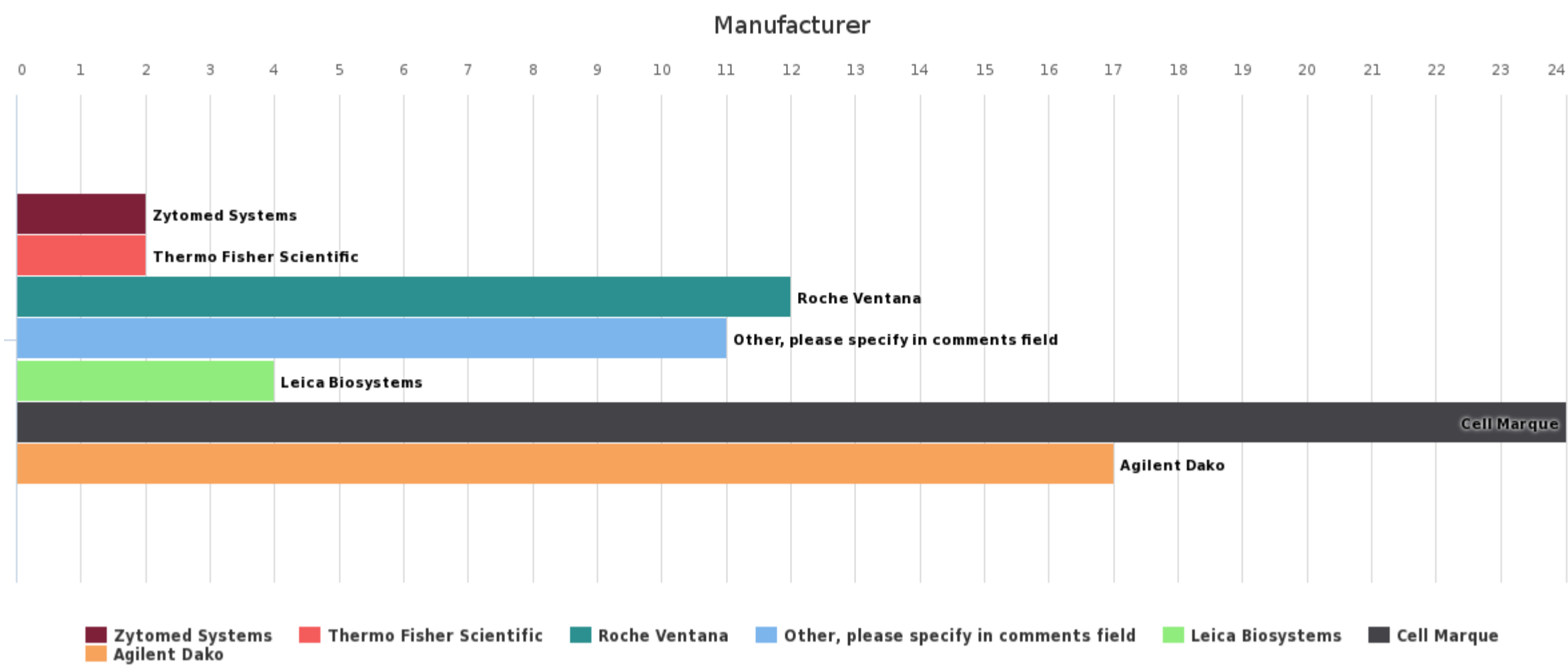


GENERAL DETAILS OF STAINING

Performance	Performance count
<input checked="" type="radio"/> Automatic	66
Manual	7
Total:	73

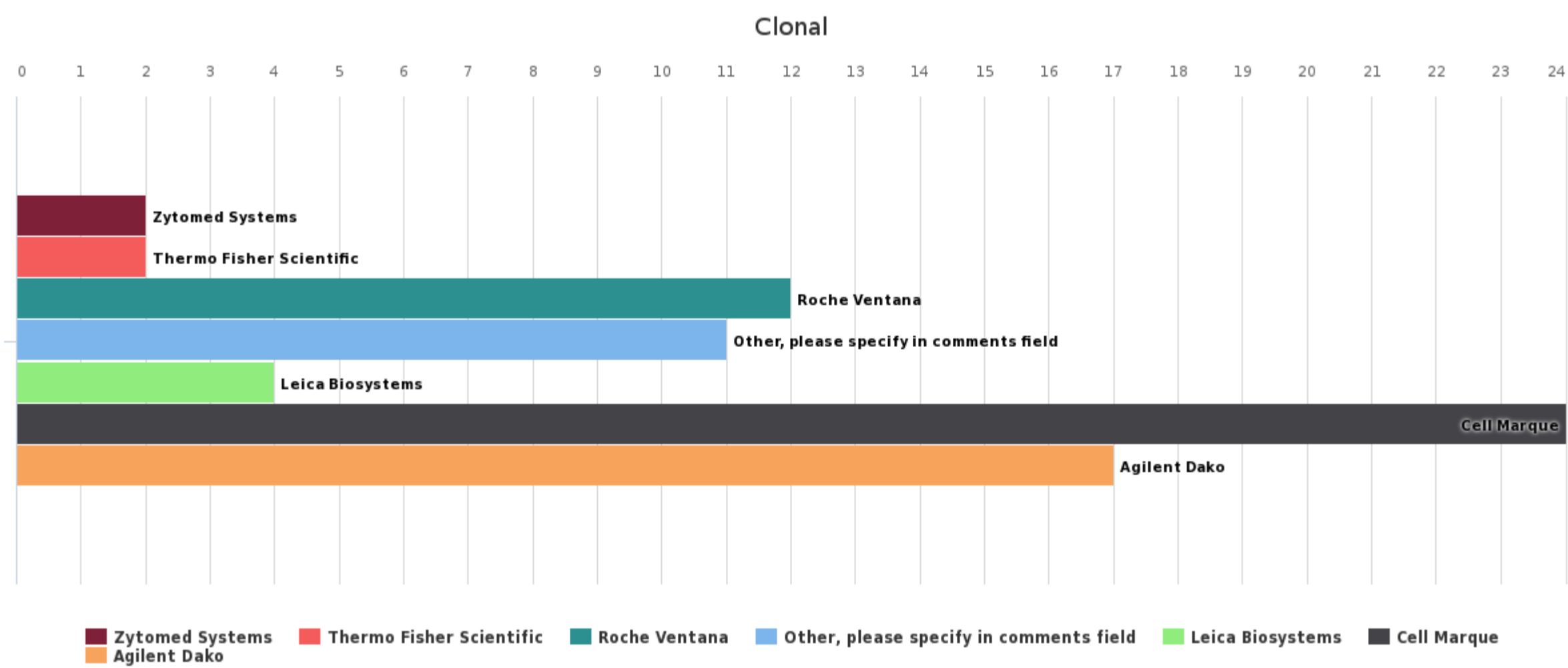


Instrument	Instrument count
Agilent Dako Autostainer Link 48	16
Agilent Dako Autostainer Plus	1
Agilent Dako Omnis	2
Leica BioSystems Bond III	5
Leica Biosystems Bondmax	6
Roche Ventana Benchmark GX	3
<input checked="" type="radio"/> Roche Ventana Benchmark Ultra	28
Roche Ventana Benchmark XT	4
Thermo Fisher Scientific Lab Vision Autostainer 480S-2D	2
Thermo Fisher Scientific Lab Vision Autostainer 720-2D	1
Total:	68

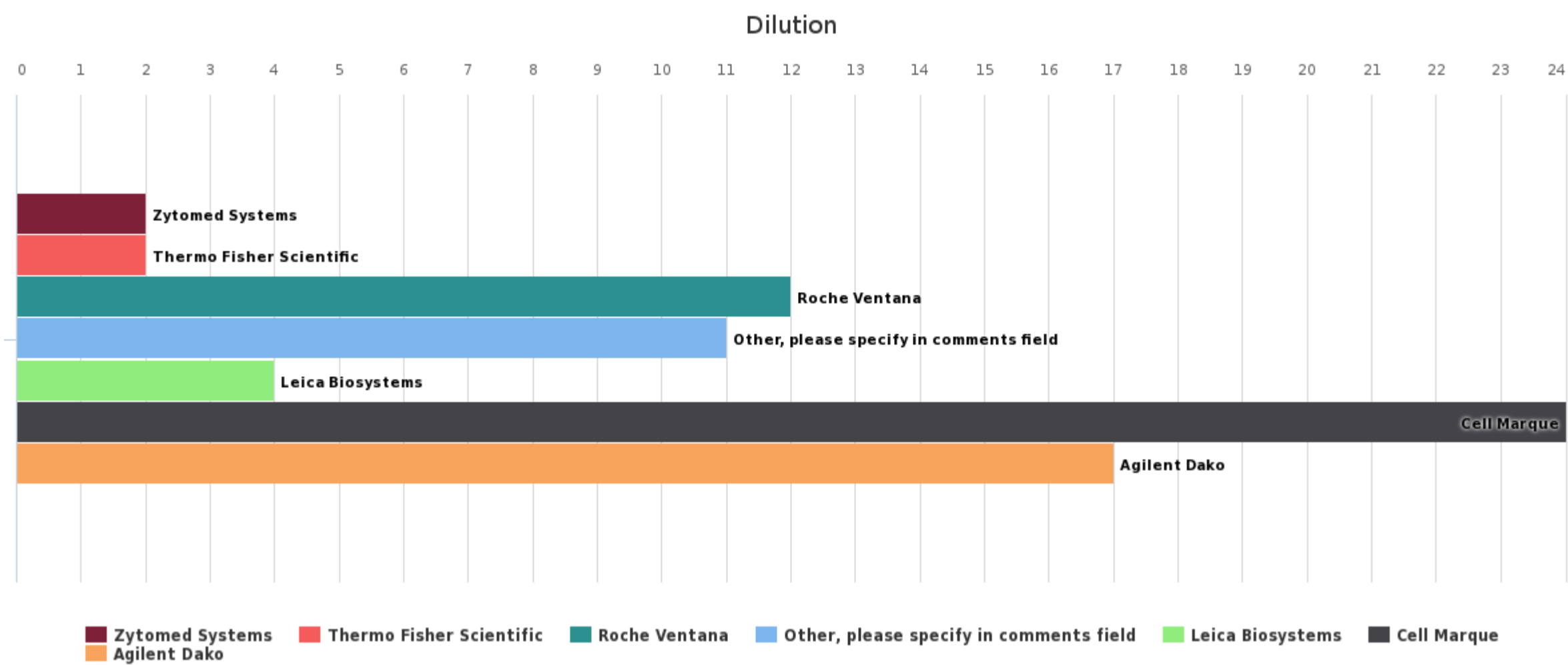


PRIMARY ANTIBODY

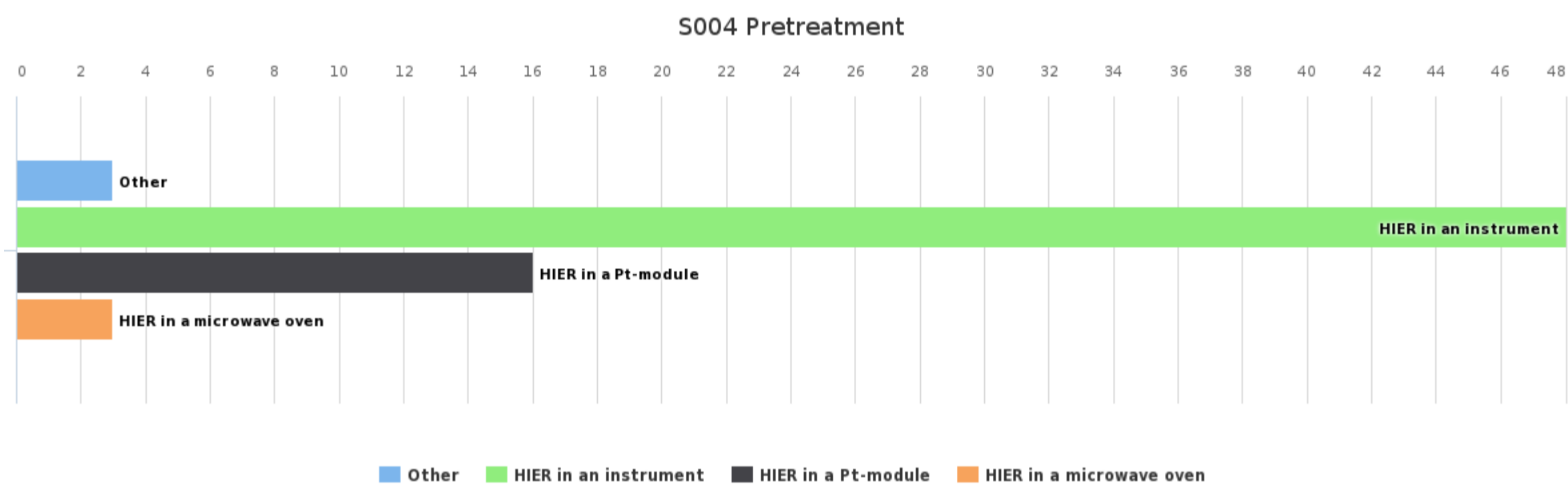
Manufacturer	Manufacturer count
Agilent Dako	17
<input checked="" type="radio"/> Cell Marque	24
Leica Biosystems	4
Other, please specify in comments field	11
Roche Ventana	12
Thermo Fisher Scientific	2
Zytomed Systems	2
Total:	72



Clonal	Clonal count
<input checked="" type="radio"/> Monoclonal	70
Polyclonal	1
Total:	71

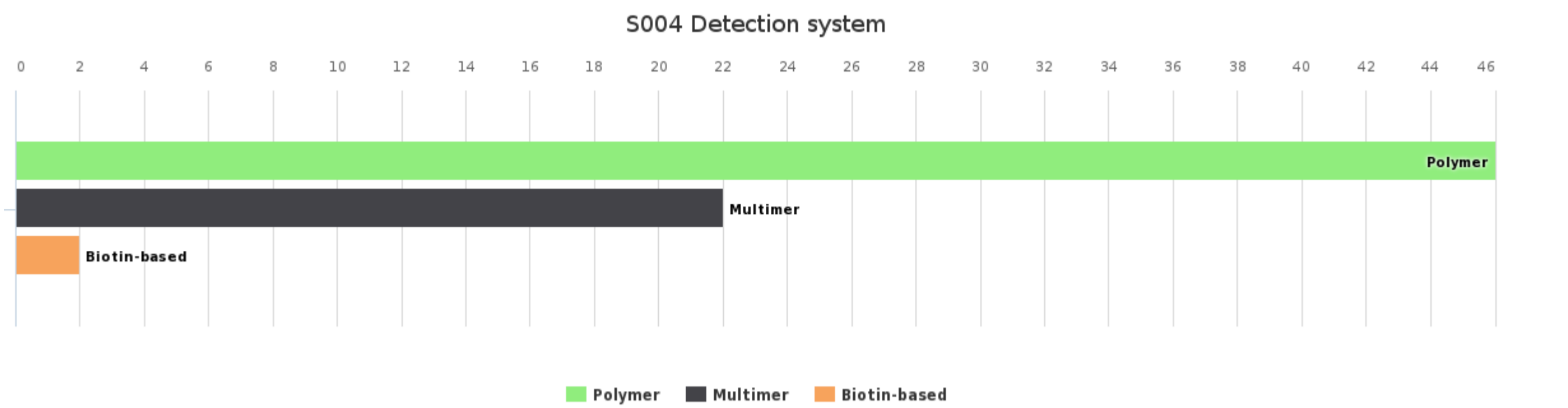


Dilution	Dilution count
Dilution	28
<input checked="" type="radio"/> Ready-to-use	44
Total:	72



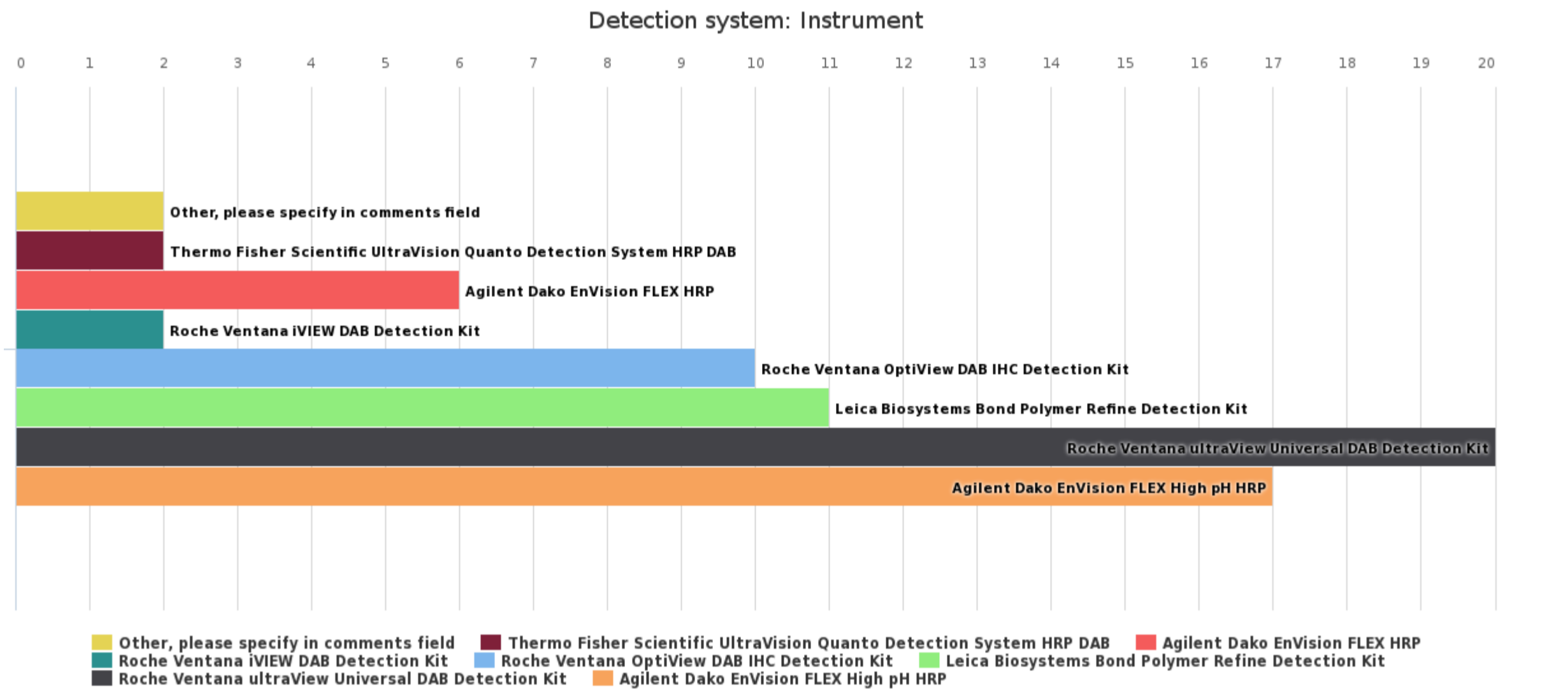
PRETREATMENT

Pretreatment	Pretreatment count
HIER in a microwave oven	3
HIER in a Pt-module	16
<input checked="" type="radio"/> HIER in an instrument	48
Other	3
Total:	70

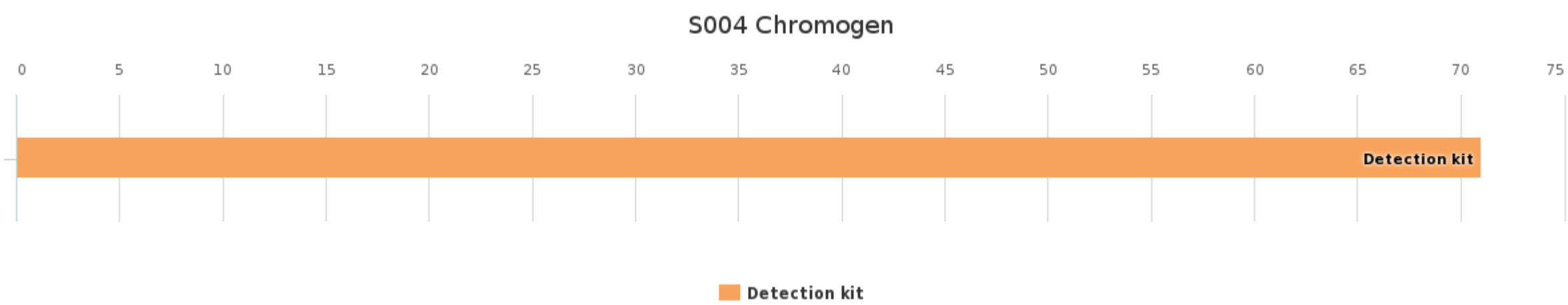


DETECTION SYSTEM

Detection system	Detection system count
Biotin-based	2
Multimer	22
<input checked="" type="radio"/> Polymer	46
Total:	70

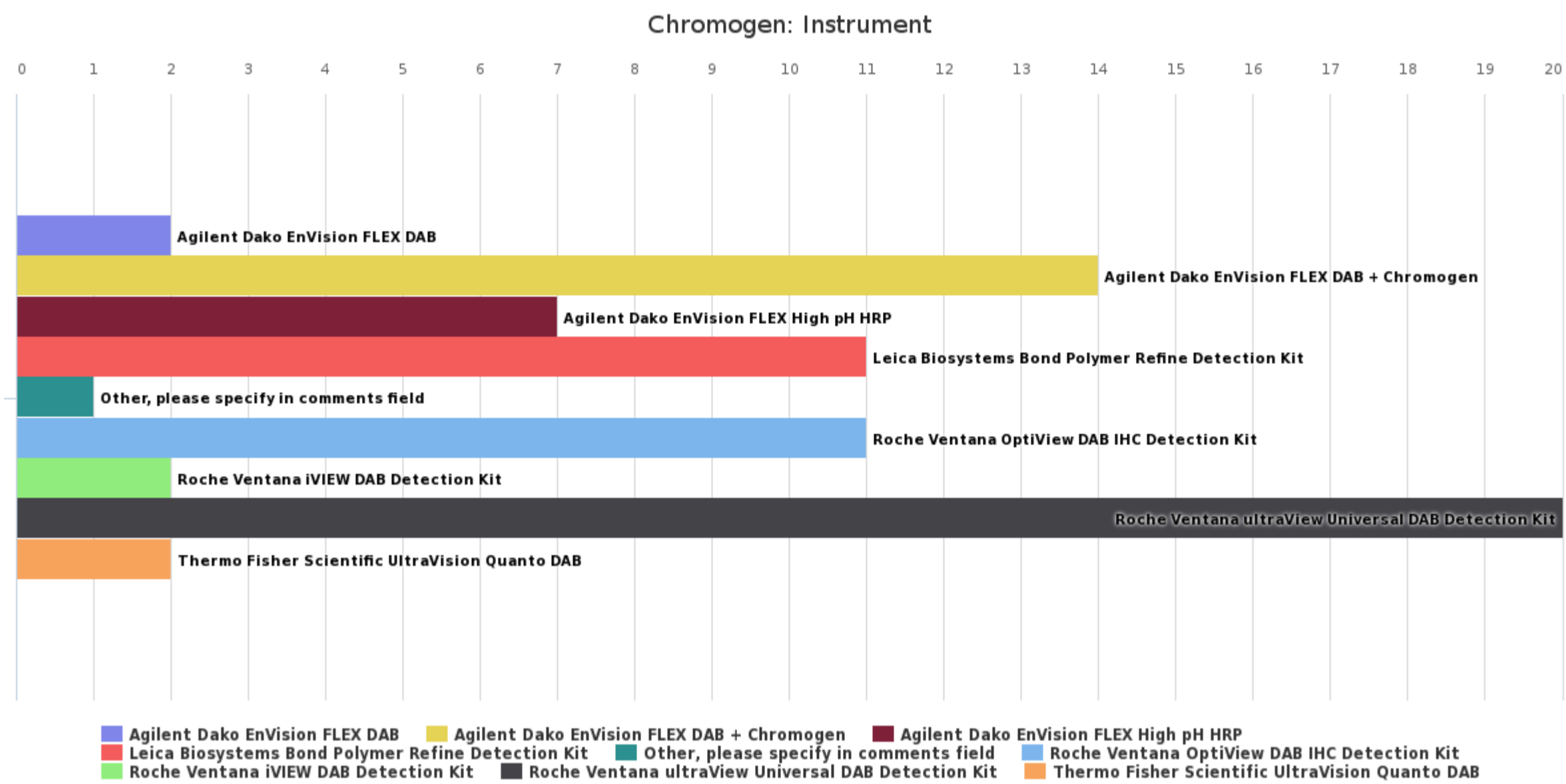


Detection system: Instrument	Detection system: Instrument count
Agilent Dako EnVision FLEX HRP	6
Agilent Dako EnVision FLEX High pH HRP	17
Leica Biosystems Bond Polymer Refine Detection Kit	11
Other, please specify in comments field	2
Roche Ventana OptiView DAB IHC Detection Kit	10
Roche Ventana iVIEW DAB Detection Kit	2
<input checked="" type="radio"/> Roche Ventana ultraView Universal DAB Detection Kit	20
Thermo Fisher Scientific UltraVision Quanto Detection System HRP DAB	2
Total:	70

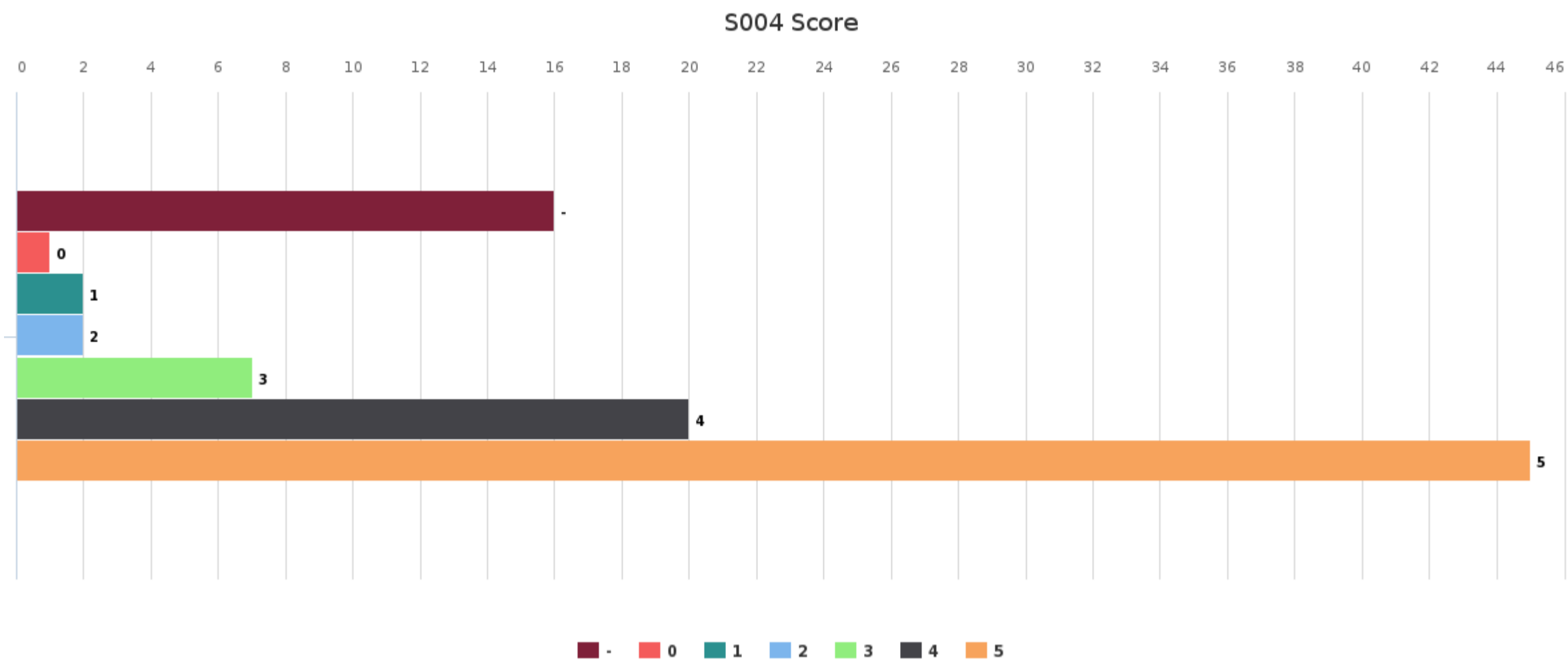


CHROMOGEN

Chromogen	Chromogen count
<input checked="" type="radio"/> Detection kit	71
Total:	71

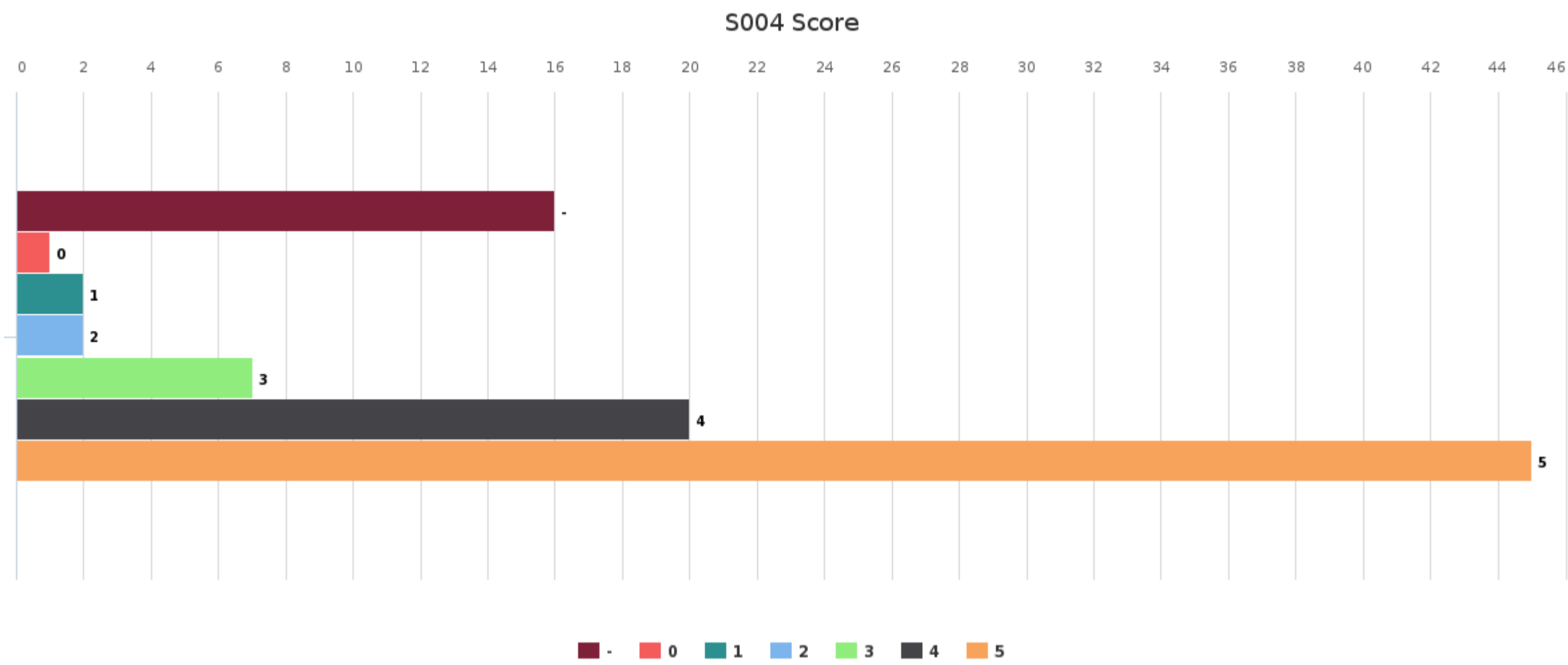


Chromogen: Instrument	Chromogen: Instrument count
Agilent Dako EnVision FLEX DAB	2
Agilent Dako EnVision FLEX DAB + Chromogen	14
Agilent Dako EnVision FLEX High pH HRP	7
Leica Biosystems Bond Polymer Refine Detection Kit	11
Other, please specify in comments field	1
Roche Ventana OptiView DAB IHC Detection Kit	11
Roche Ventana iVIEW DAB Detection Kit	2
<input checked="" type="radio"/> Roche Ventana ultraView Universal DAB Detection Kit	20
Thermo Fisher Scientific UltraVision Quanto DAB	2
Total:	70



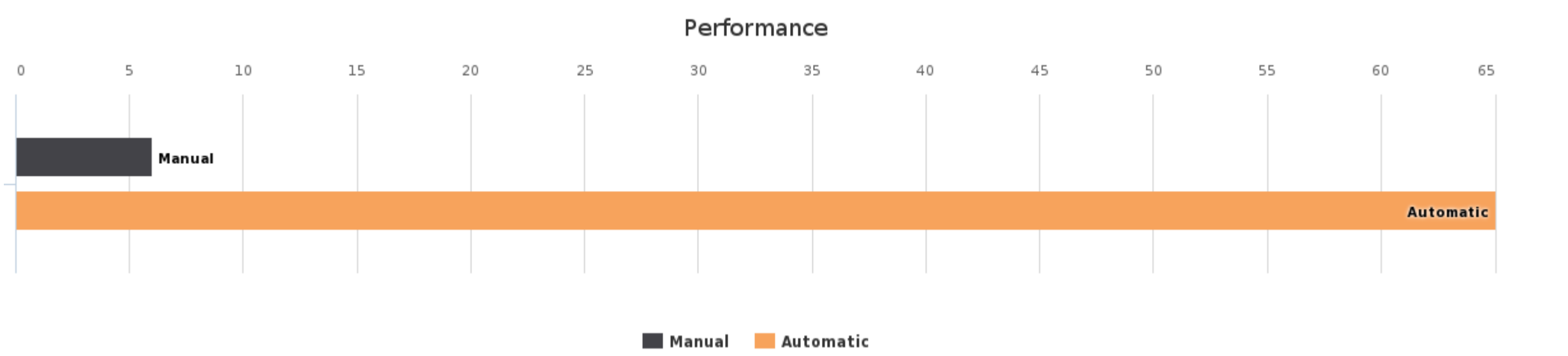
SCORE

Score	Comment
-	not available
0	false negative
1	almost negative staining, inadequate staining very weak staining, almost negative
2	understaining, unspecific staining weak staining
3	stain deposits, uneven, understained understained understained, uneven undertained, some background staining
4	dark hematoxylin, hematoxylin deposits slightly overstained slightly understained slightly understained, hematoxylin deposits slightly understained, stain deposits slightly understained, uneven slightly uneven some background staining stain deposits uneven, hematoxylin deposits uneven, some background staining uneven, stain deposits
<input checked="" type="radio"/> 5	<input checked="" type="radio"/> Excellent hematoxylin deposits
Total:	



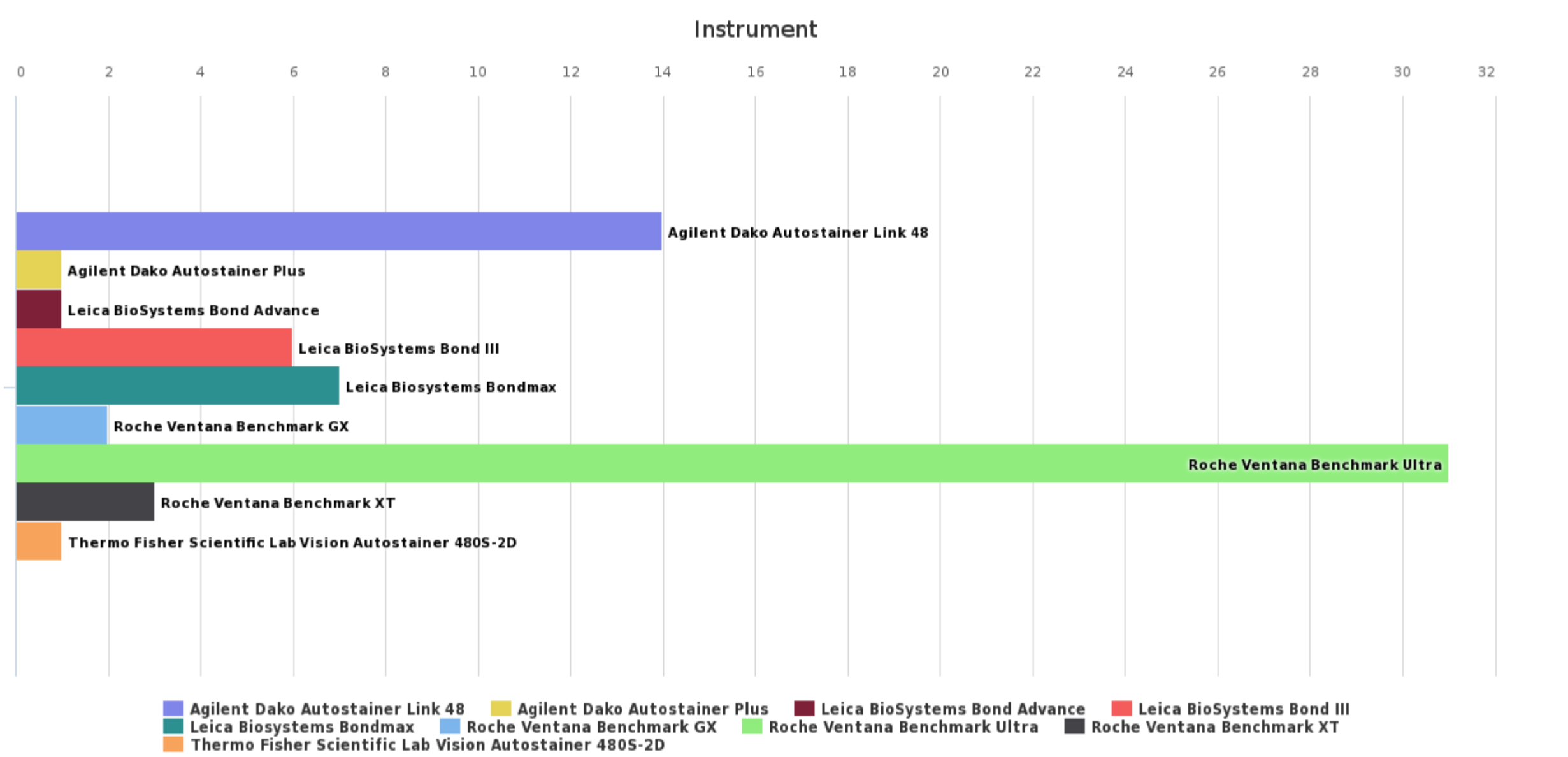
Score	Score count
-	16
0	1
1	2
2	2
3	7
4	20
<input checked="" type="radio"/> 5	45
Total:	93

S005 | Sample S005 PSA

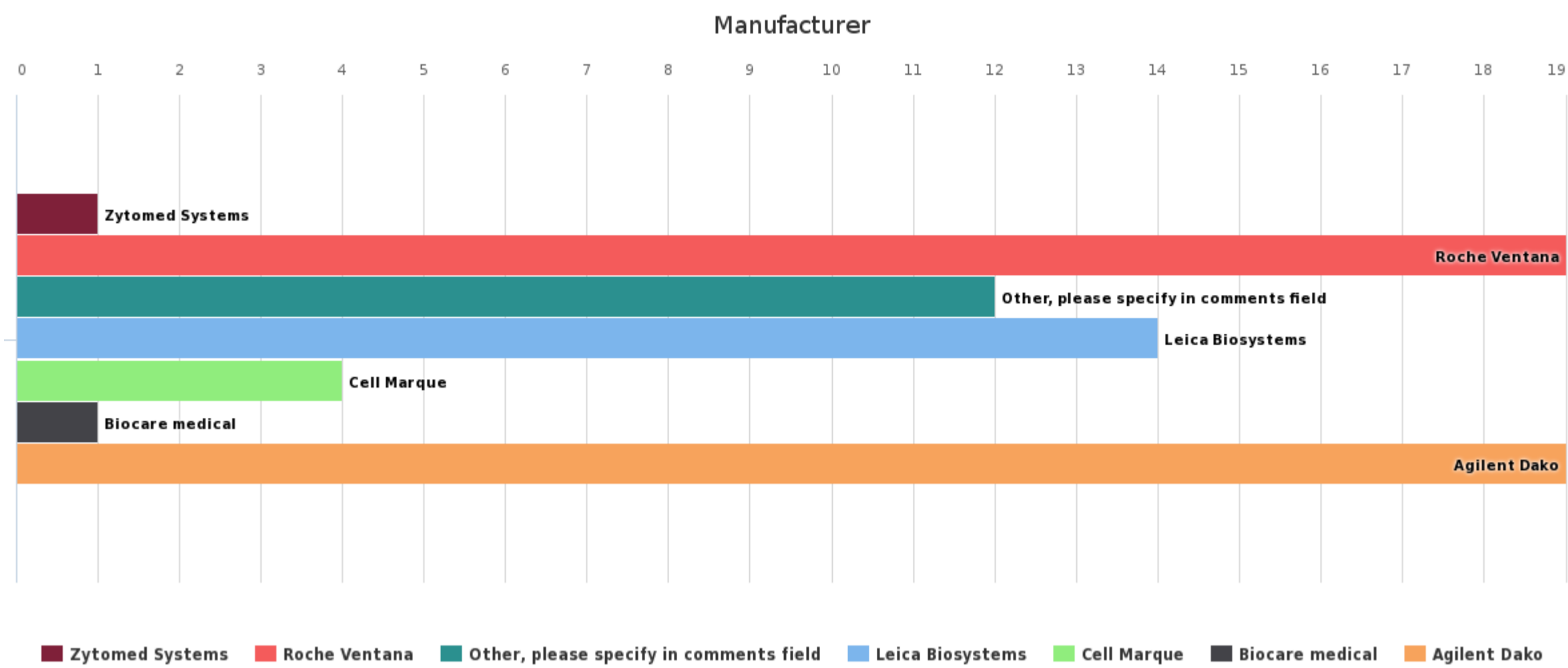


GENERAL DETAILS OF STAINING

Performance	Performance count
<input checked="" type="radio"/> Automatic	65
Manual	6
Total:	71

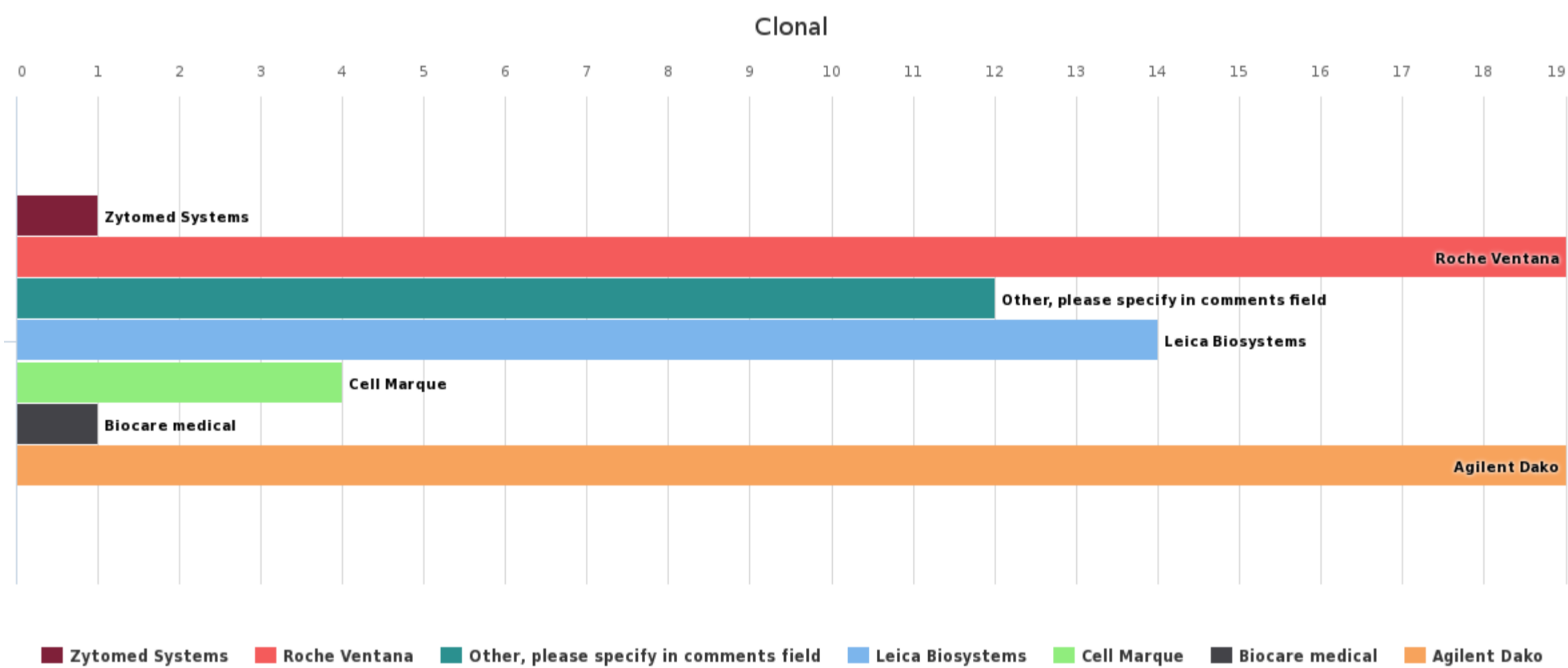


Instrument	Instrument count
Agilent Dako Autostainer Link 48	14
Agilent Dako Autostainer Plus	1
Leica BioSystems Bond Advance	1
Leica BioSystems Bond III	6
Leica Biosystems Bondmax	7
Roche Ventana Benchmark GX	2
<input checked="" type="radio"/> Roche Ventana Benchmark Ultra	31
Roche Ventana Benchmark XT	3
Thermo Fisher Scientific Lab Vision Autostainer 480S-2D	1
Total:	66

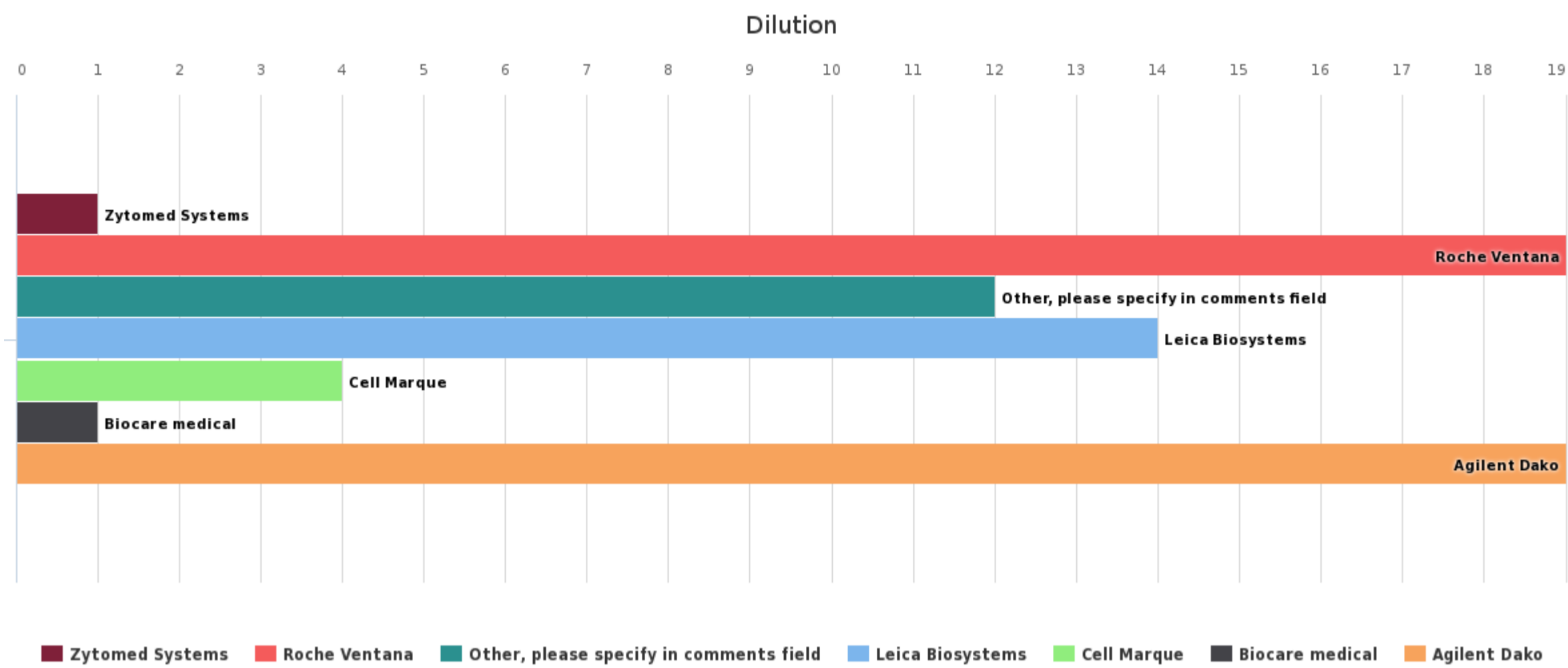


PRIMARY ANTIBODY

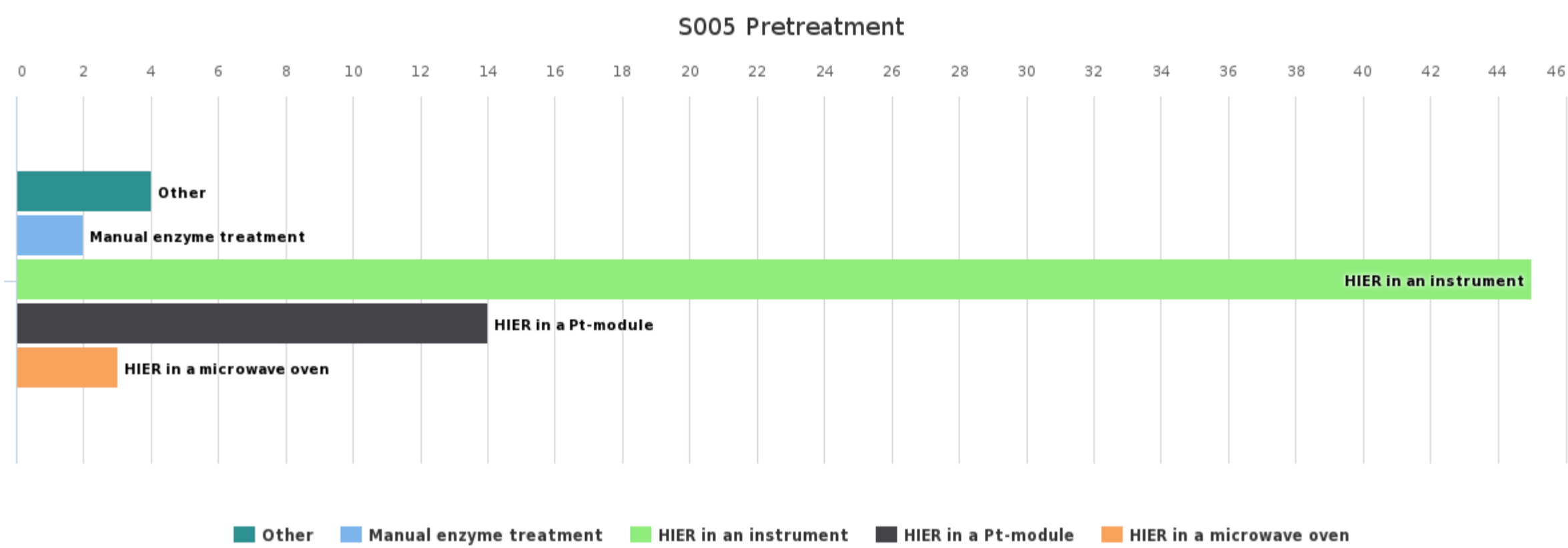
Manufacturer	Manufacturer count
Agilent Dako	19
Biocare medical	1
Cell Marque	4
Leica Biosystems	14
Other, please specify in comments field	12
<input checked="" type="radio"/> Roche Ventana	19
Zytomed Systems	1
Total:	70



Clonal	Clonal count
<input checked="" type="radio"/> Monoclonal	50
Polyclonal	19
Total:	69

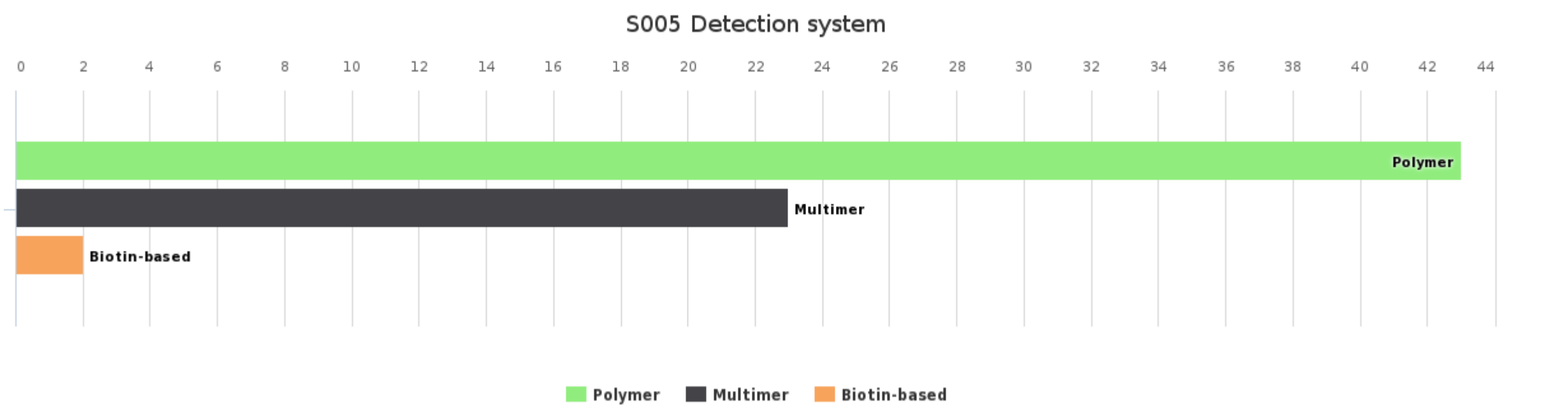


Dilution	Dilution count
Dilution	40
<input checked="" type="radio"/> Ready-to-use	30
Total:	70



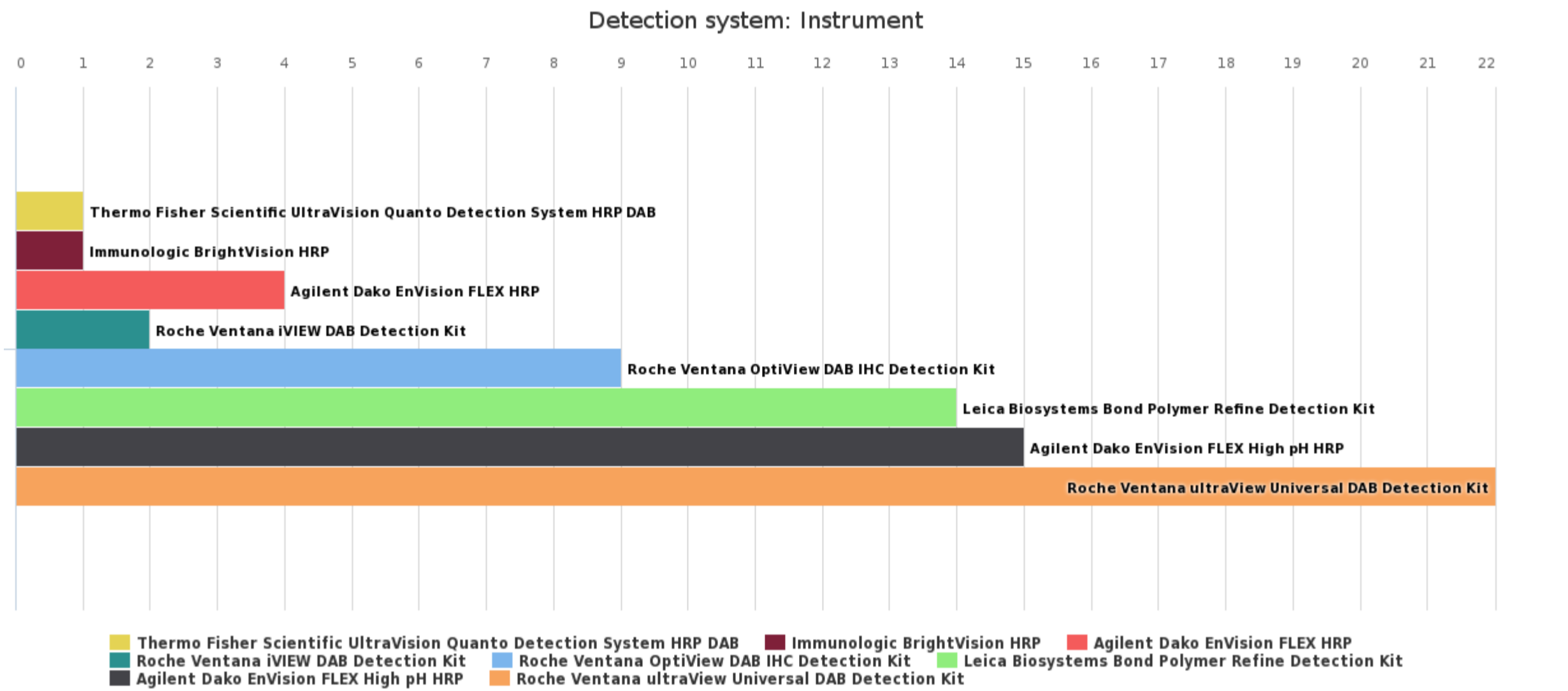
PRETREATMENT

Pretreatment	Pretreatment count
HIER in a microwave oven	3
HIER in a Pt-module	14
<input checked="" type="radio"/> HIER in an instrument	45
Manual enzyme treatment	2
Other	4
Total:	68

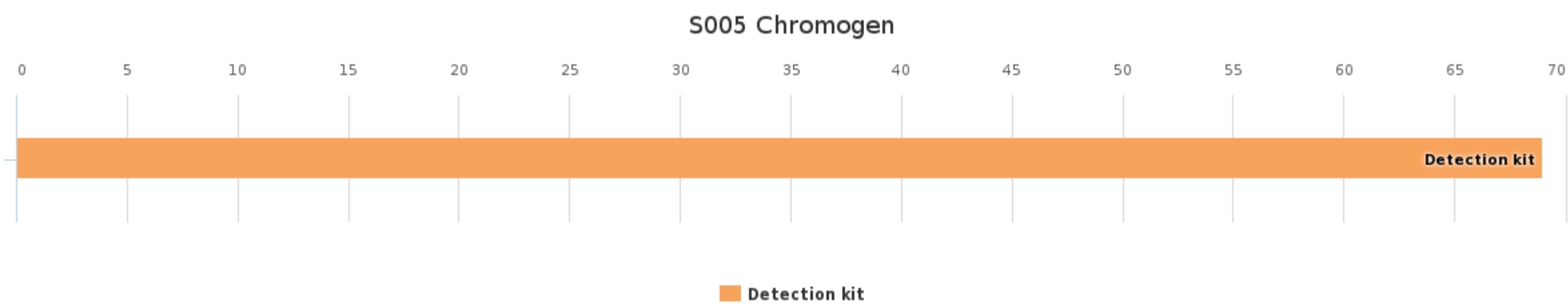


DETECTION SYSTEM

Detection system	Detection system count
Biotin-based	2
Multimer	23
<input checked="" type="radio"/> Polymer	43
Total:	68

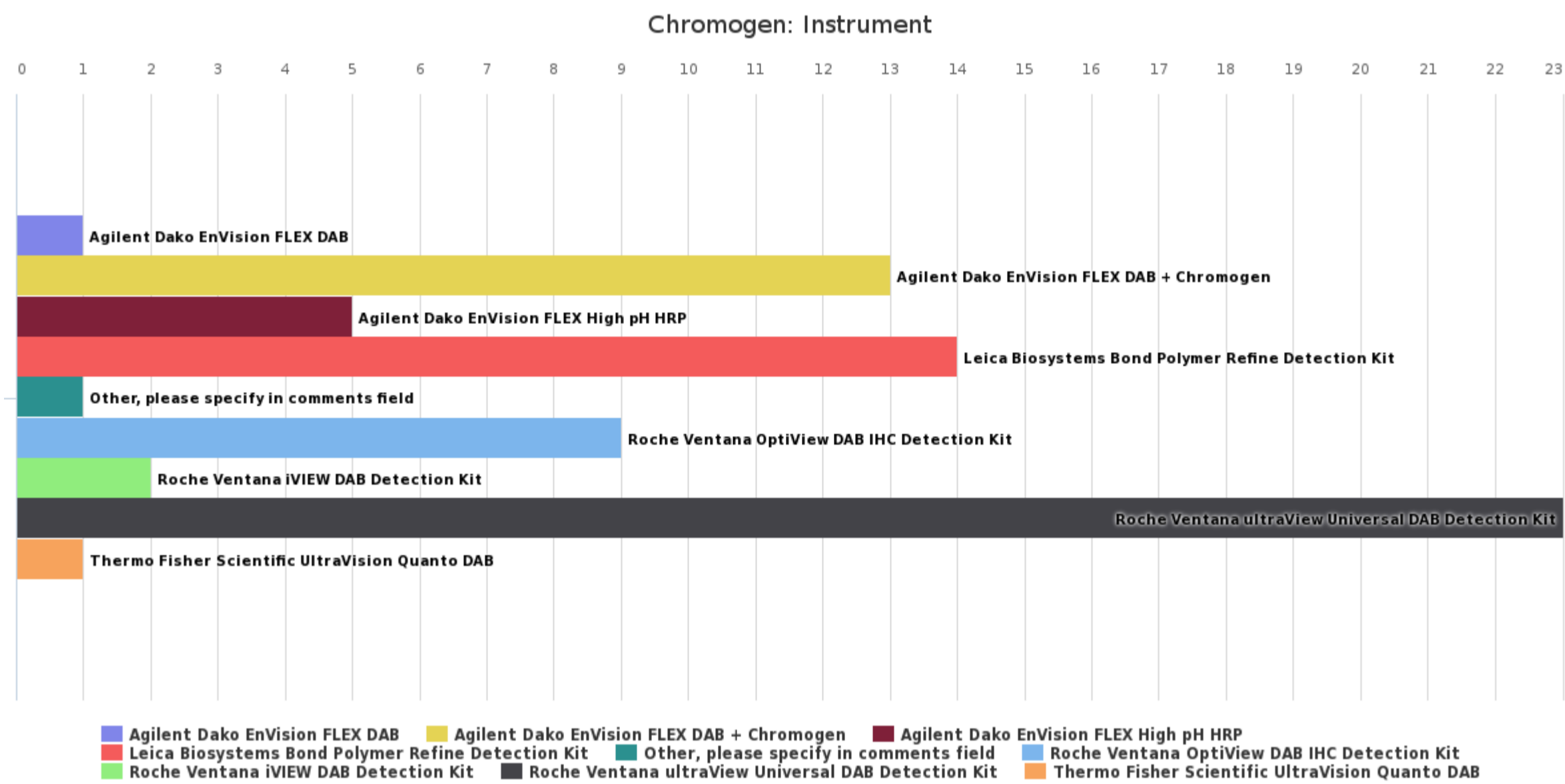


Detection system: Instrument	Detection system: Instrument count
Agilent Dako EnVision FLEX HRP	4
Agilent Dako EnVision FLEX High pH HRP	15
Immunologic BrightVision HRP	1
Leica Biosystems Bond Polymer Refine Detection Kit	14
Roche Ventana OptiView DAB IHC Detection Kit	9
Roche Ventana iVIEW DAB Detection Kit	2
<input checked="" type="radio"/> Roche Ventana ultraView Universal DAB Detection Kit	22
Thermo Fisher Scientific UltraVision Quanto Detection System HRP DAB	1
Total:	68

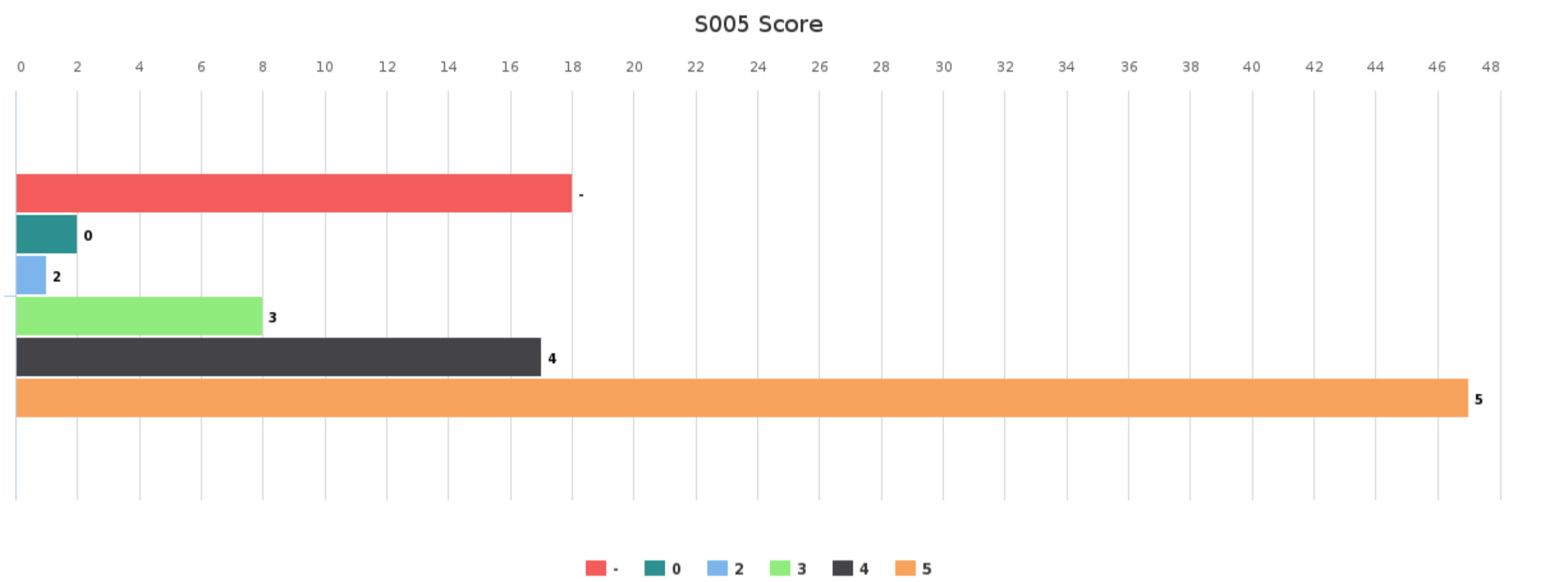


CHROMOGEN

Chromogen	Chromogen count
<input checked="" type="radio"/> Detection kit	69
Total:	69

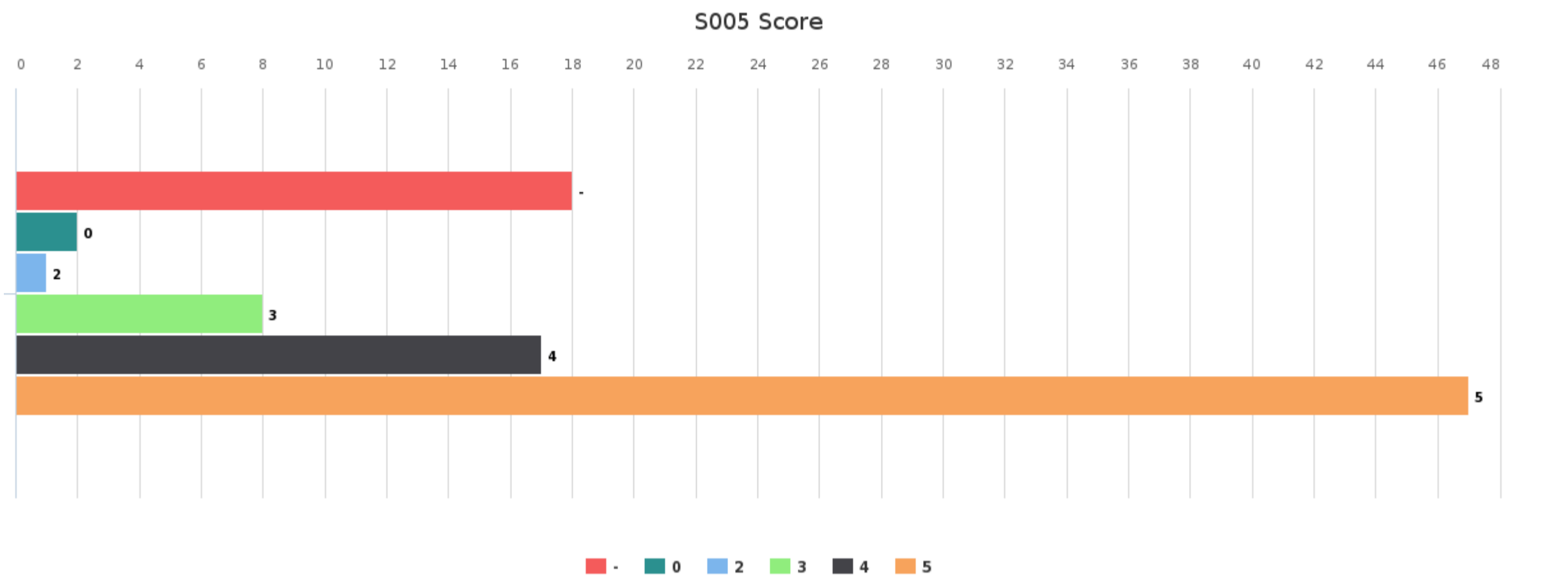


Chromogen: Instrument	Chromogen: Instrument count
Agilent Dako EnVision FLEX DAB	1
Agilent Dako EnVision FLEX DAB + Chromogen	13
Agilent Dako EnVision FLEX High pH HRP	5
Leica Biosystems Bond Polymer Refine Detection Kit	14
Other, please specify in comments field	1
Roche Ventana OptiView DAB IHC Detection Kit	9
Roche Ventana iVIEW DAB Detection Kit	2
<input checked="" type="radio"/> Roche Ventana ultraView Universal DAB Detection Kit	23
Thermo Fisher Scientific UltraVision Quanto DAB	1
Total:	69



SCORE

Score	Comment
-	
	not available
0	
	negative
2	
	weak staining
3	
	dark hematoxylin, understained
	understained
	understained, dark hematoxylin
	understained, hematoxylin is quite weak
4	
	dark hematoxylin
	slightly understained
	slightly understained and uneven
	slightly understained, uneven
	slightly uneven
	uneven
<input checked="" type="radio"/> 5	
	<input checked="" type="radio"/> Excellent
Total:	

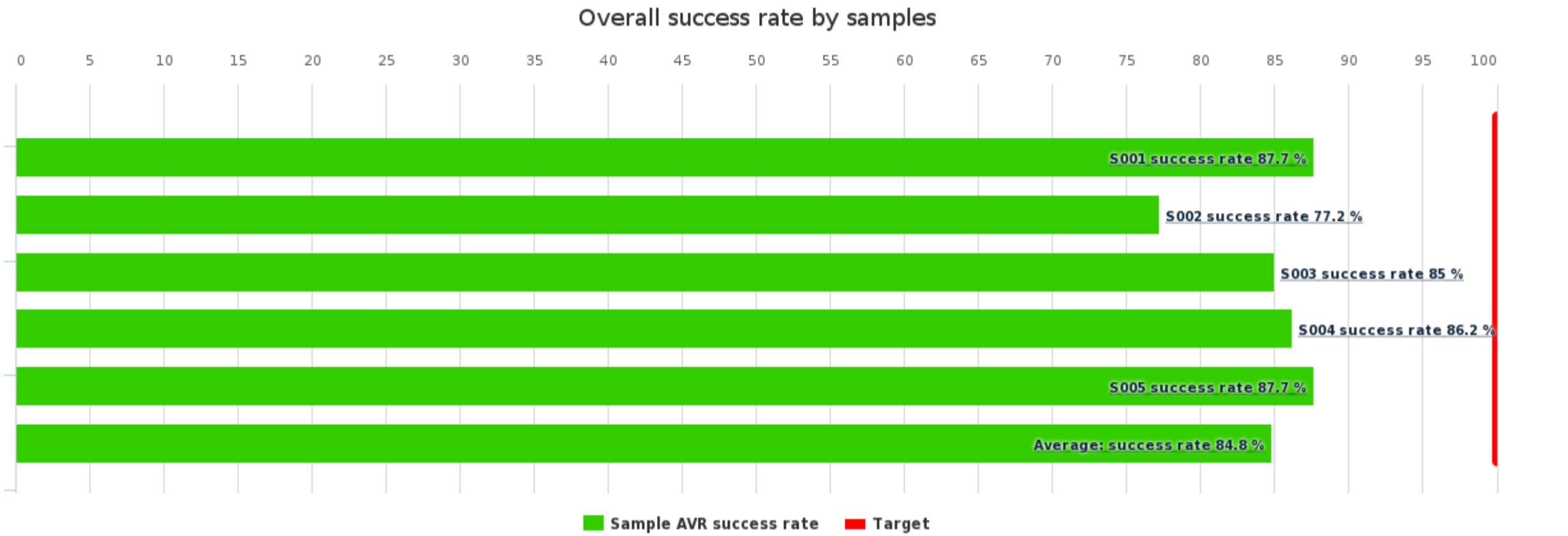


Score	Score count
-	18
0	2
2	1
3	8
4	17
<input checked="" type="radio"/> 5	47
Total:	93

GLOBAL REPORT

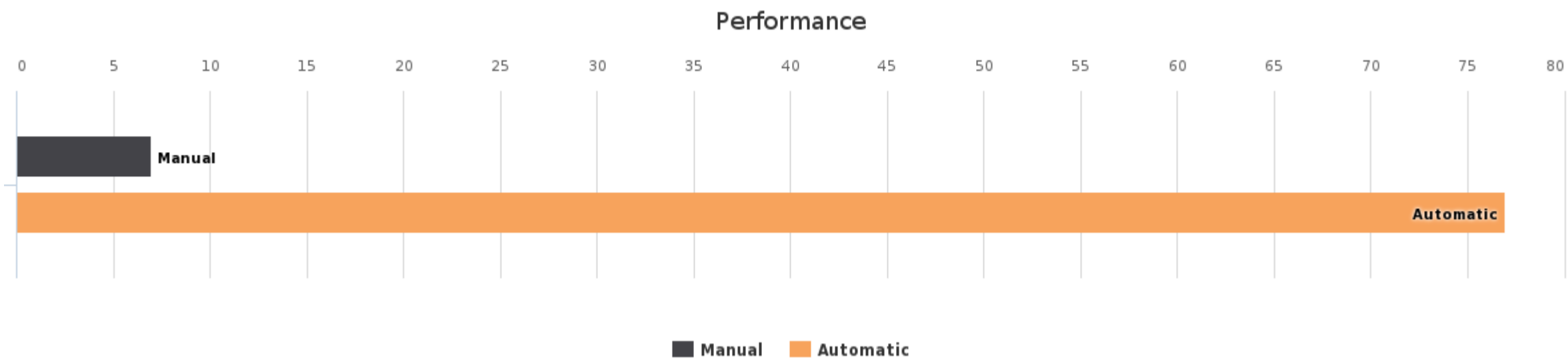
	No of participants	No of responded participants	Response percentage
Immunohistochemical staining methods, limited selection of antibodies, November, 3-2022	29	29	100 %
Immunohistochemical staining methods, November, 3-2022	65	65	100 %

Summary



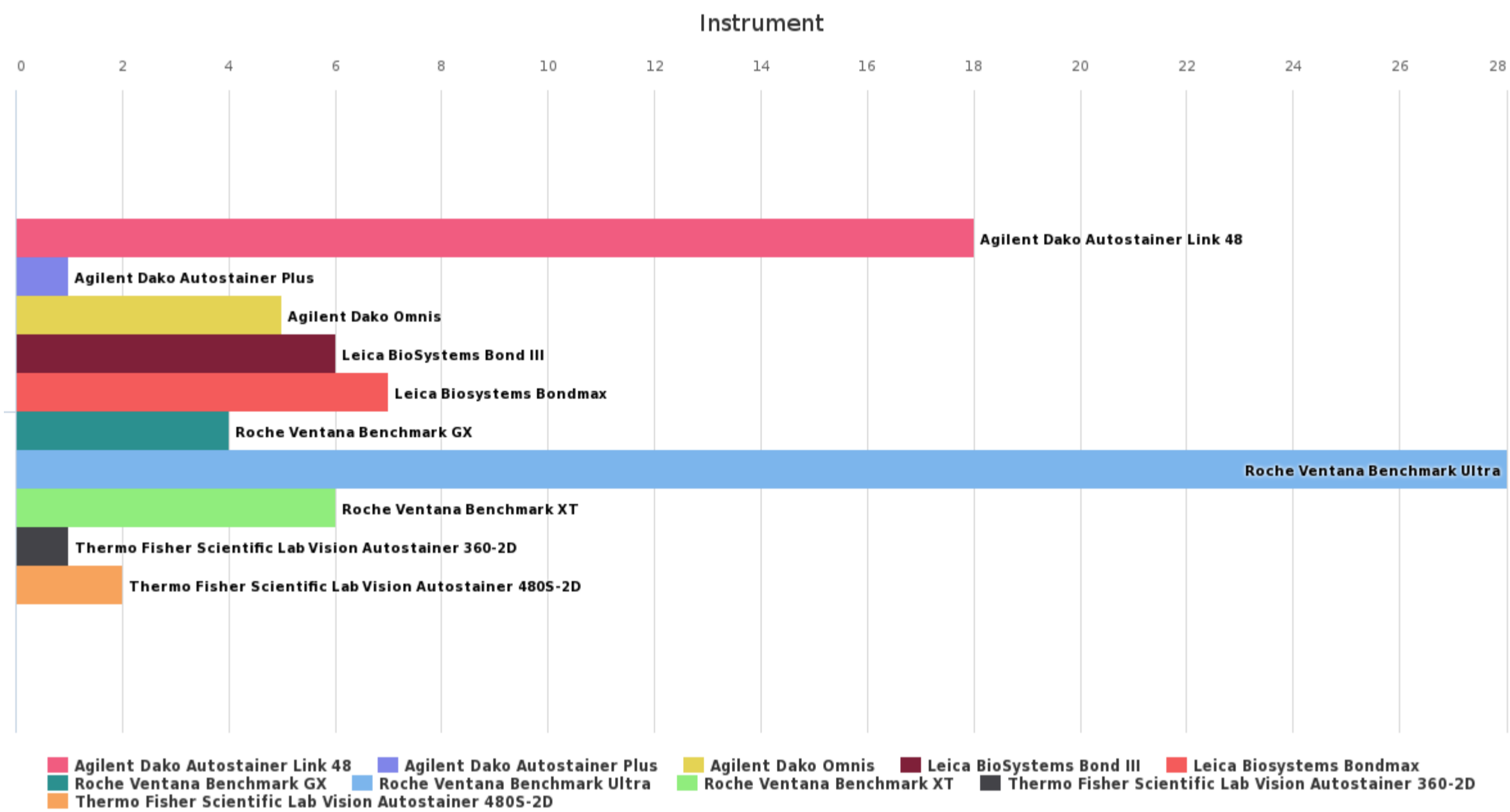
Summary	AVR success rate
S001	87.7 %
S002	77.2 %
S003	85 %
S004	86.2 %
S005	87.7 %
Average:	84.8 %

S001 | Sample S001 CK7

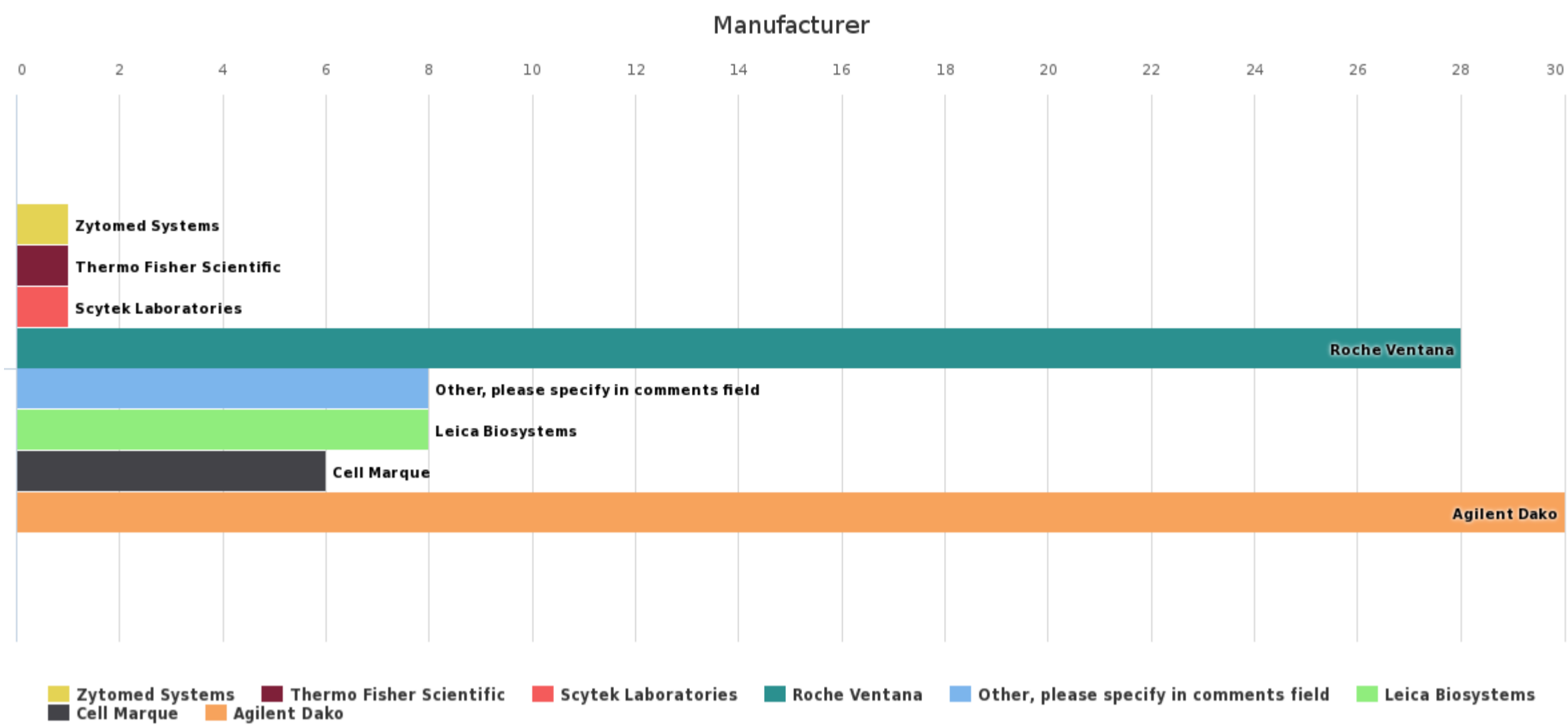


GENERAL DETAILS OF STAINING

Performance	Performance count
Automatic	77
Manual	7
Total:	84

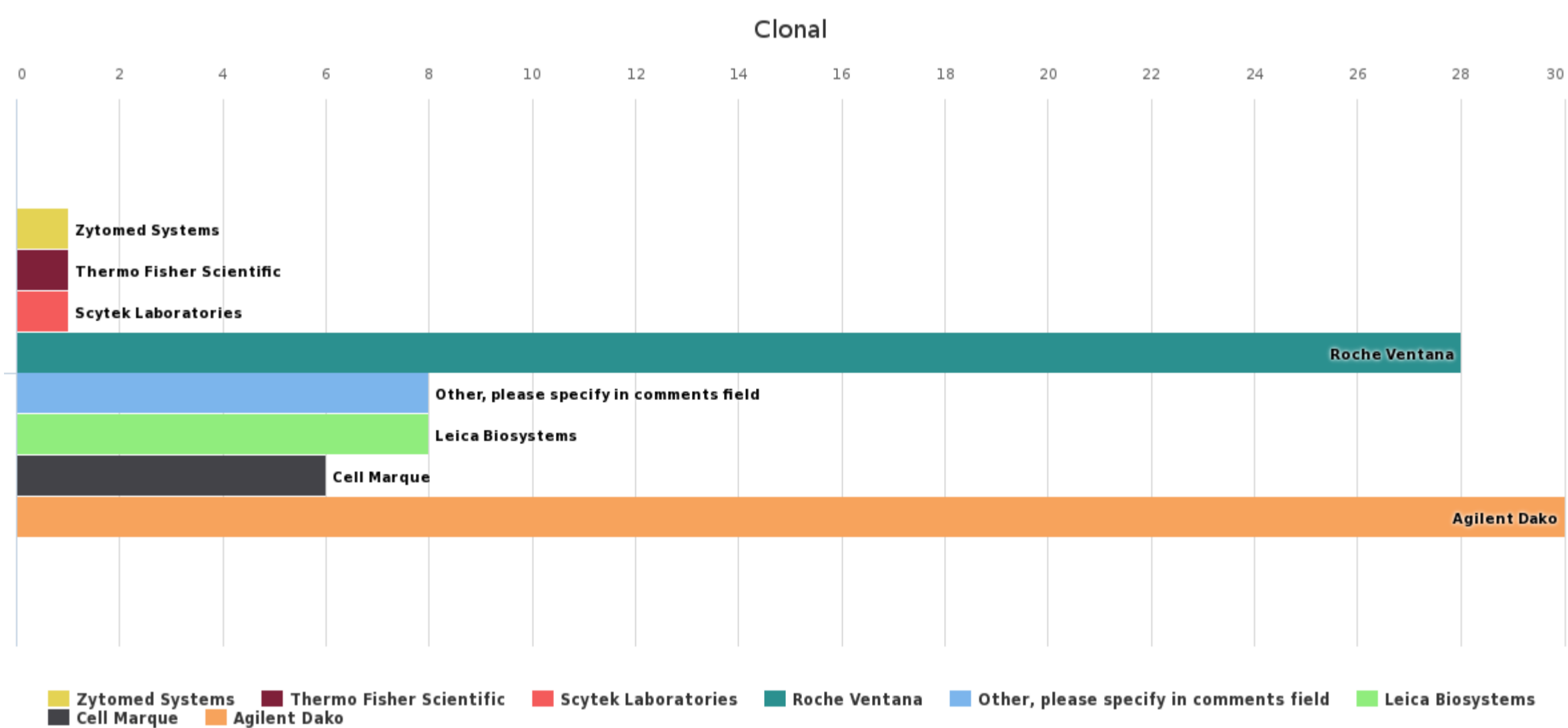


Instrument	Instrument count
Agilent Dako Autostainer Link 48	18
Agilent Dako Autostainer Plus	1
Agilent Dako Omnis	5
Leica BioSystems Bond III	6
Leica Biosystems Bondmax	7
Roche Ventana Benchmark GX	4
Roche Ventana Benchmark Ultra	28
Roche Ventana Benchmark XT	6
Thermo Fisher Scientific Lab Vision Autostainer 360-2D	1
Thermo Fisher Scientific Lab Vision Autostainer 480S-2D	2
Total:	78

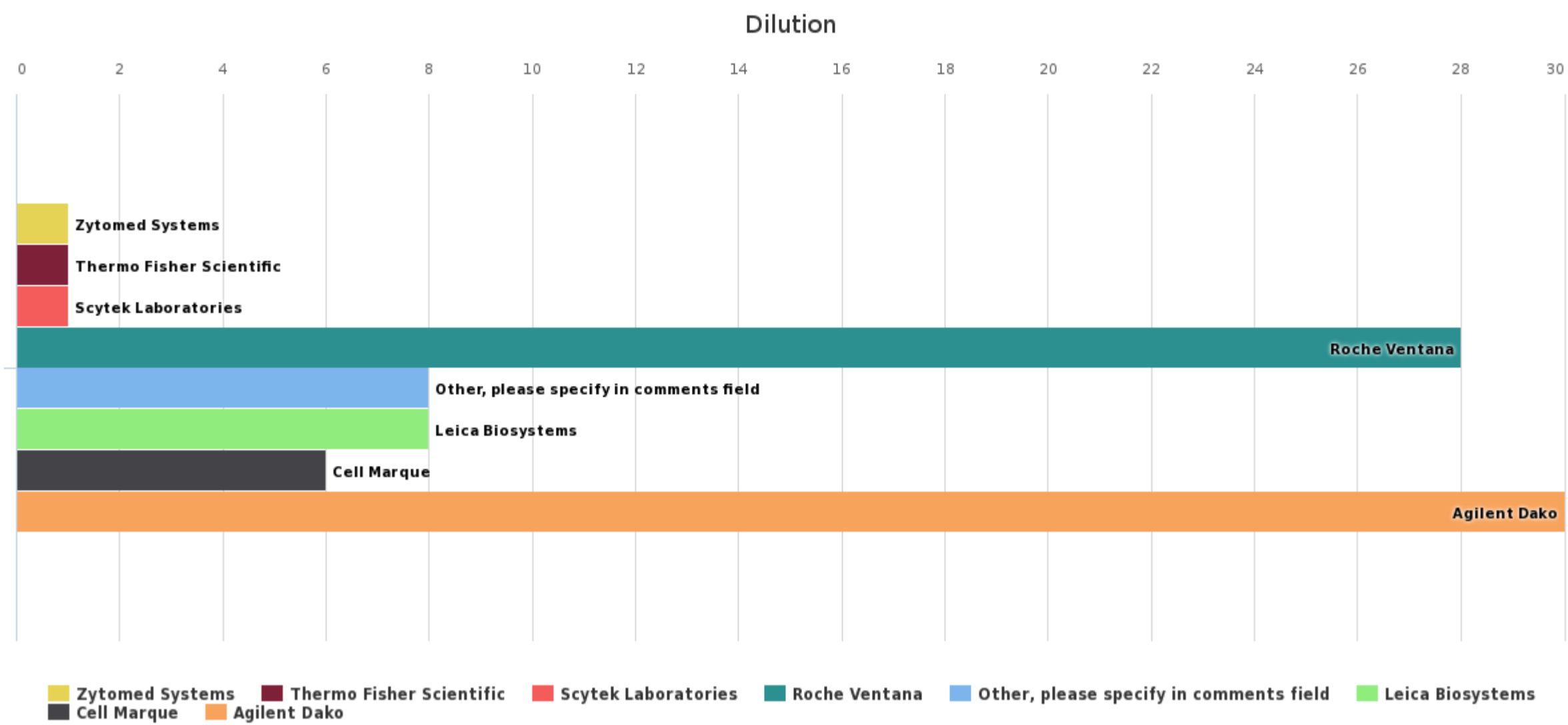


PRIMARY ANTIBODY

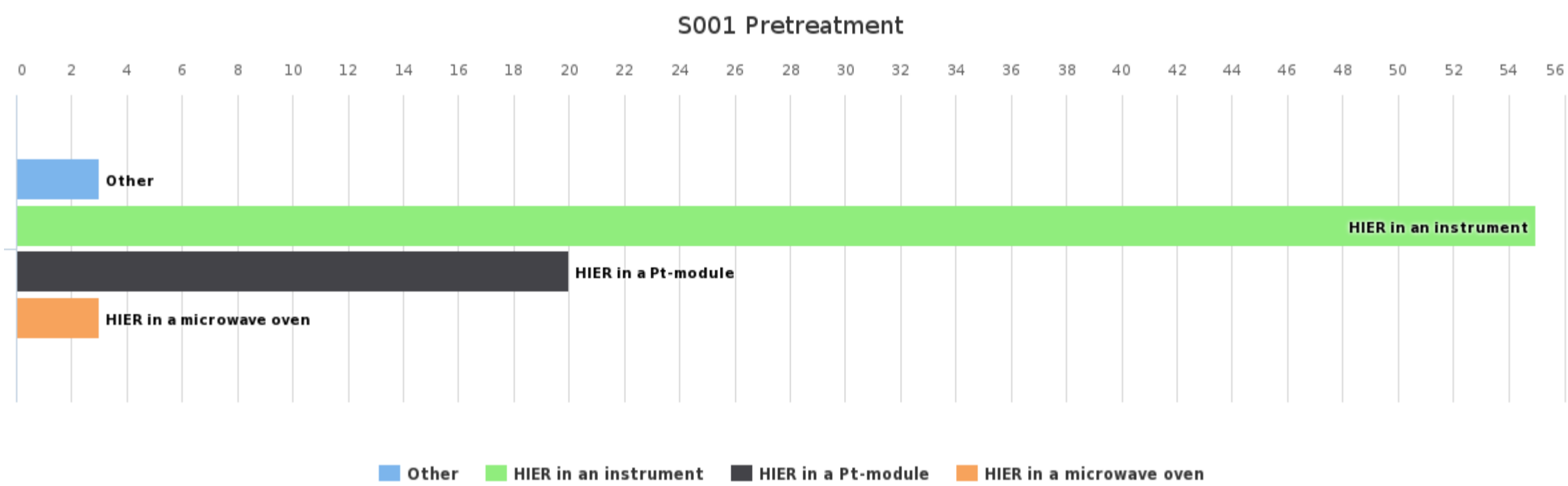
Manufacturer	Manufacturer count
Agilent Dako	30
Cell Marque	6
Leica Biosystems	8
Other, please specify in comments field	8
Roche Ventana	28
Scytek Laboratories	1
Thermo Fisher Scientific	1
Zytomed Systems	1
Total:	83



Clonal	Clonal count
Monoclonal	82
Total:	82

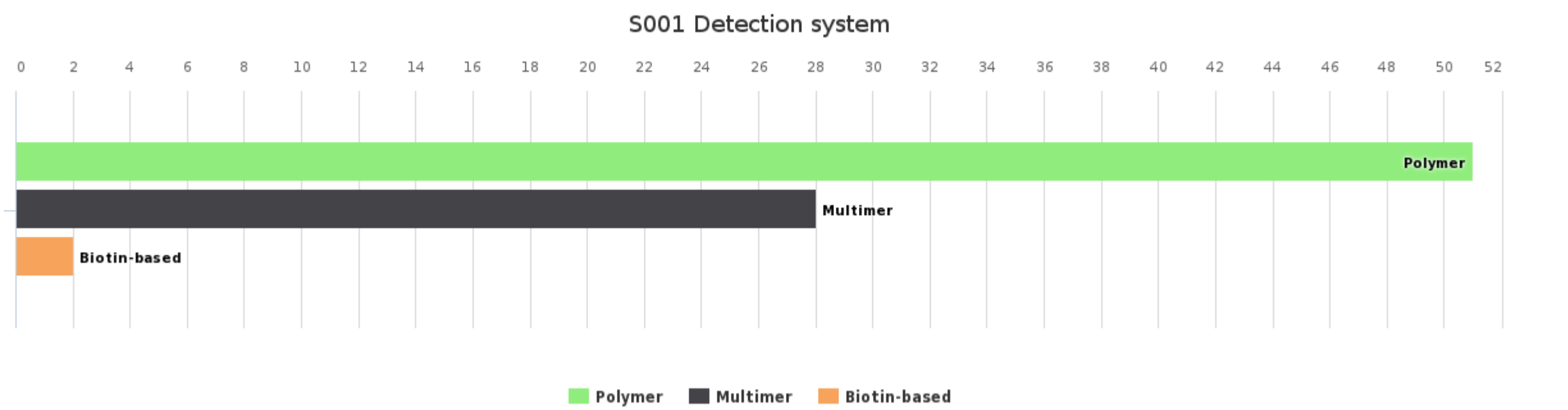


Dilution	Dilution count
Dilution	33
Ready-to-use	50
Total:	83



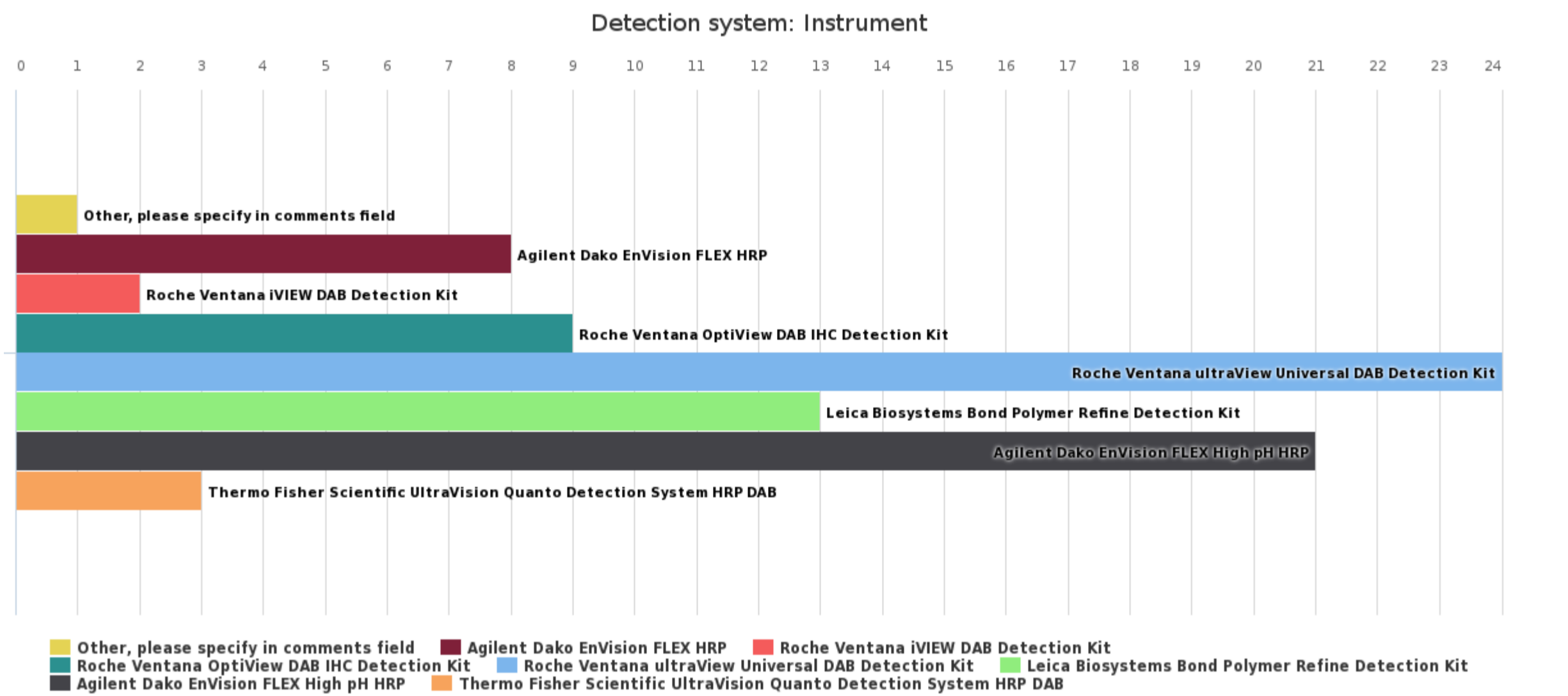
PRETREATMENT

Pretreatment	Pretreatment count
HIER in a microwave oven	3
HIER in a Pt-module	20
HIER in an instrument	55
Other	3
Total:	81

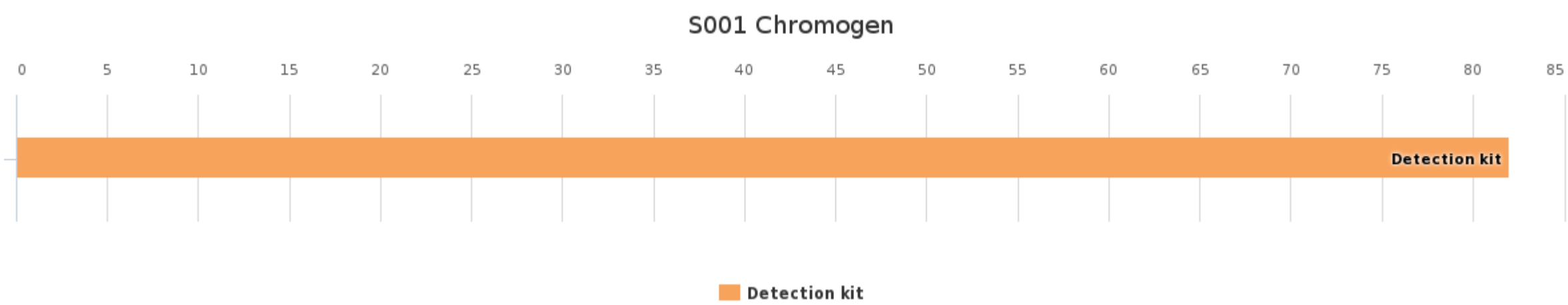


DETECTION SYSTEM

Detection system	Detection system count
Biotin-based	2
Multimer	28
Polymer	51
Total:	81

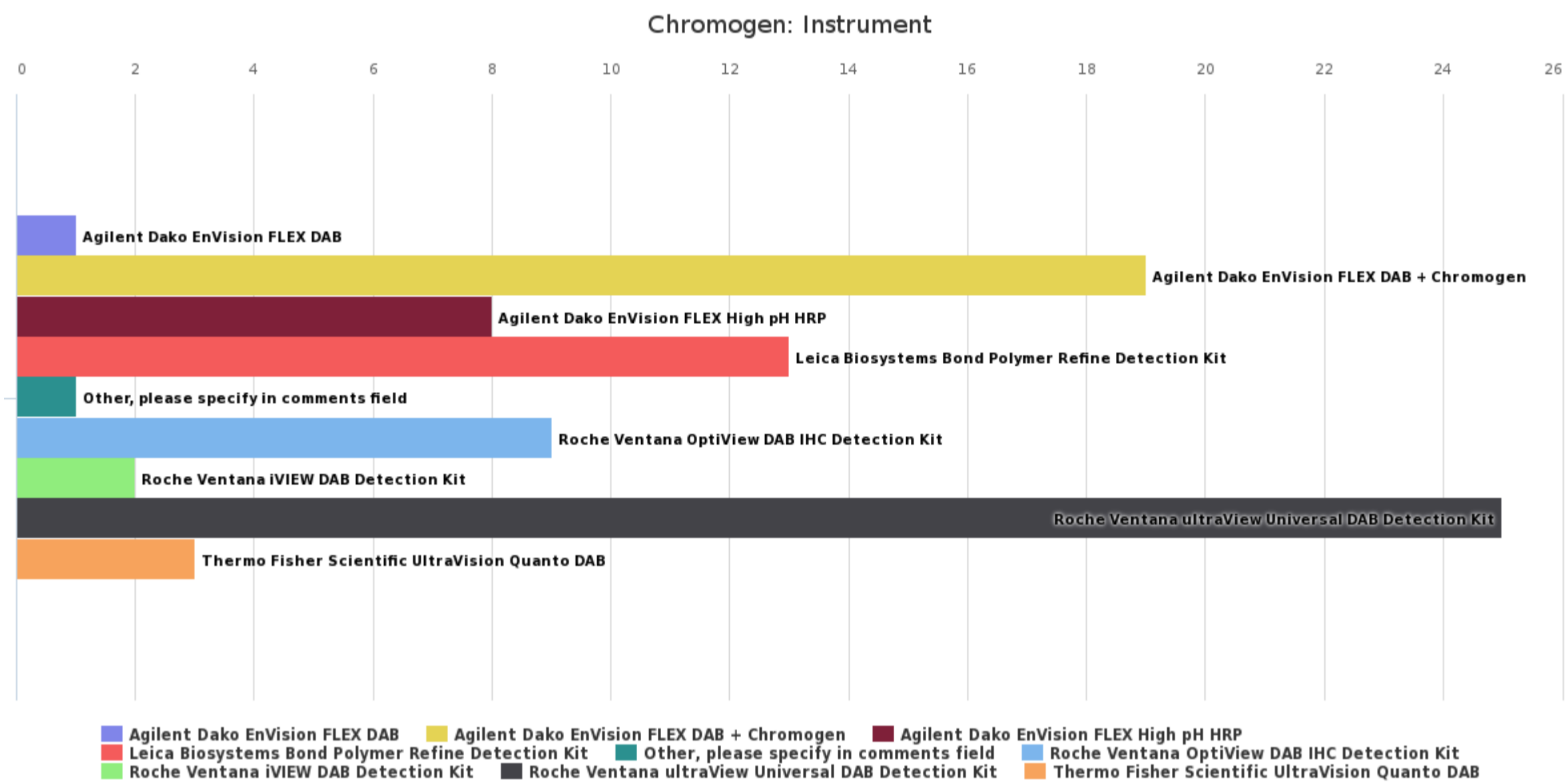


Detection system: Instrument	Detection system: Instrument count
Agilent Dako EnVision FLEX HRP	8
Agilent Dako EnVision FLEX High pH HRP	21
Leica Biosystems Bond Polymer Refine Detection Kit	13
Other, please specify in comments field	1
Roche Ventana OptiView DAB IHC Detection Kit	9
Roche Ventana iVIEW DAB Detection Kit	2
Roche Ventana ultraView Universal DAB Detection Kit	24
Thermo Fisher Scientific UltraVision Quanto Detection System HRP DAB	3
Total:	81

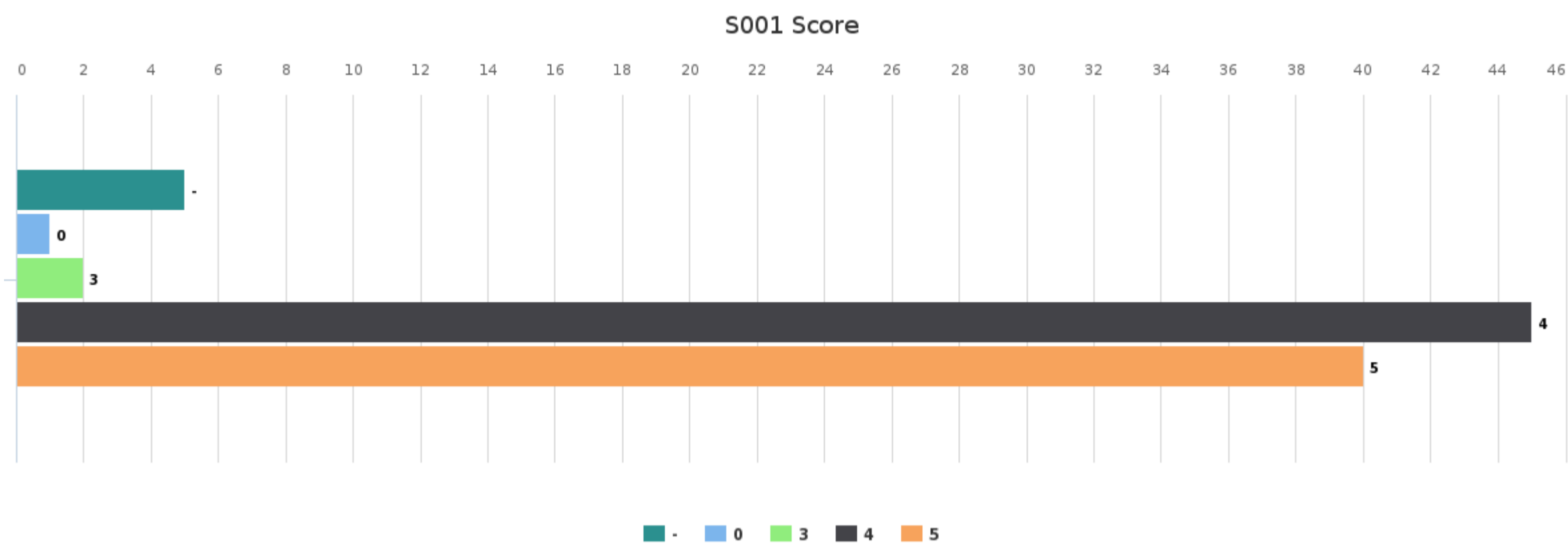


CHROMOGEN

Chromogen	Chromogen count
Detection kit	82
Total:	82

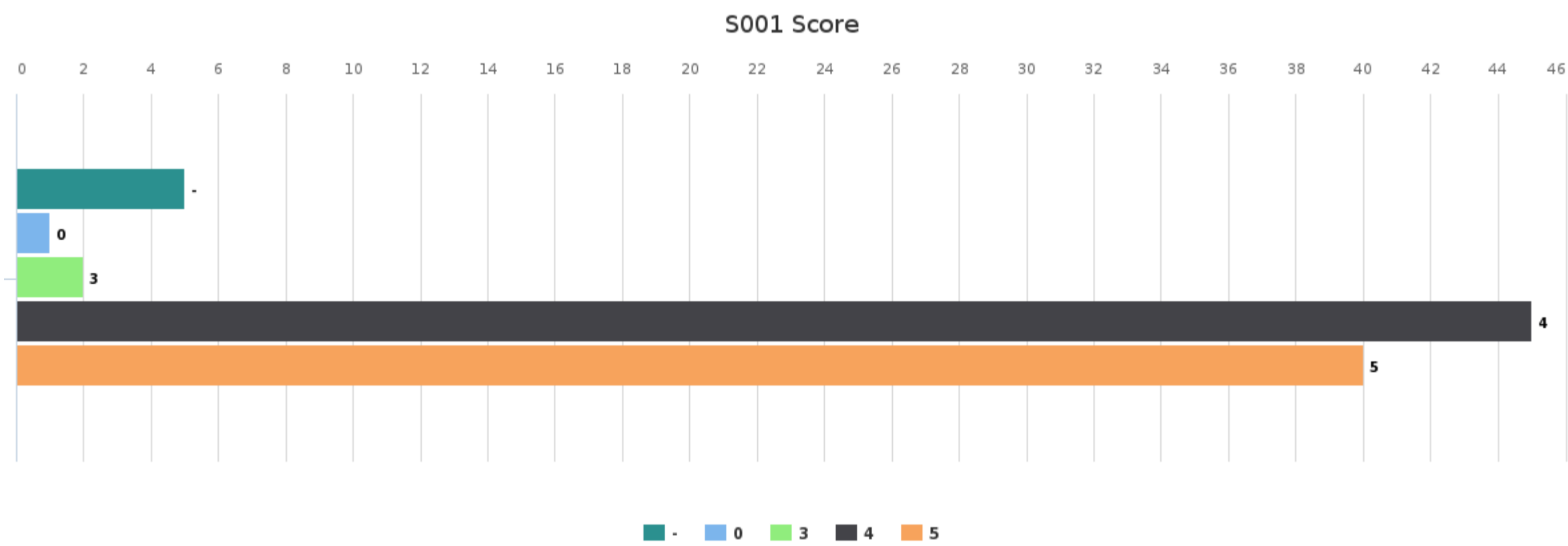


Chromogen: Instrument	Chromogen: Instrument count
Agilent Dako EnVision FLEX DAB	1
Agilent Dako EnVision FLEX DAB + Chromogen	19
Agilent Dako EnVision FLEX High pH HRP	8
Leica Biosystems Bond Polymer Refine Detection Kit	13
Other, please specify in comments field	1
Roche Ventana OptiView DAB IHC Detection Kit	9
Roche Ventana iVIEW DAB Detection Kit	2
Roche Ventana ultraView Universal DAB Detection Kit	25
Thermo Fisher Scientific UltraVision Quanto DAB	3
Total:	81



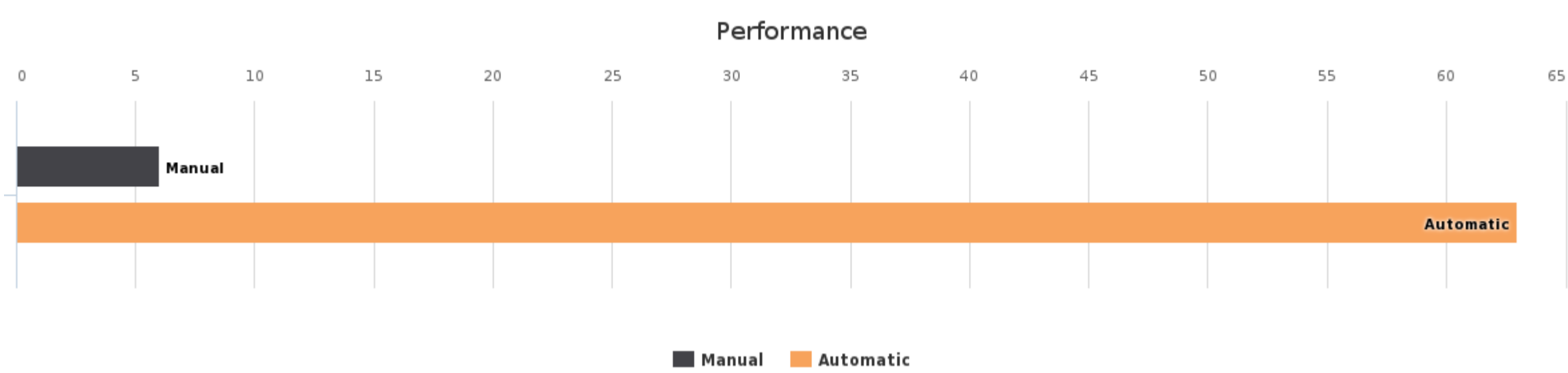
SCORE

Score	Comment
-	
	not available
0	
	negative
3	
	one core is not stained, technical issue?
	uneven, some background staining, one core is missing
4	
	dark hematoxylin
	slightly understained
	slightly understained, uneven
	slightly uneven
	some background staining
	some background staining ,slighly uneven
	some background staining, dark hematoxylin
	some background staining, hematoxylin is quite weak
	some background staining, slightly overstained
	some background staining, slightly uneven
	some background, hematoxylin in pancreas sample is uneven
	some background staining
5	
	dark hematoxylin
	Excellent
Total:	



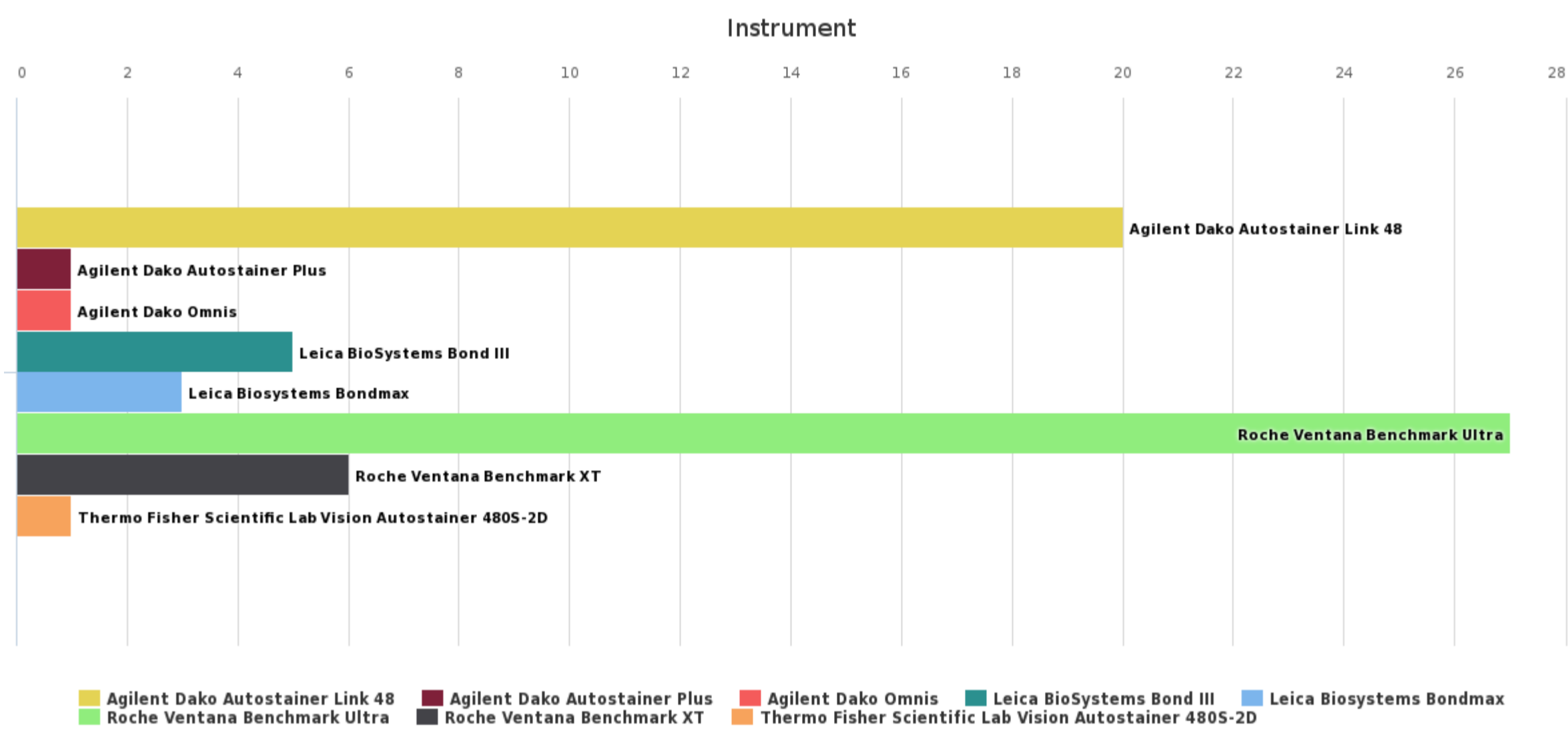
Score	Score count
-	5
0	1
3	2
4	45
5	40
Total:	93

S002 | Sample S002 WT1

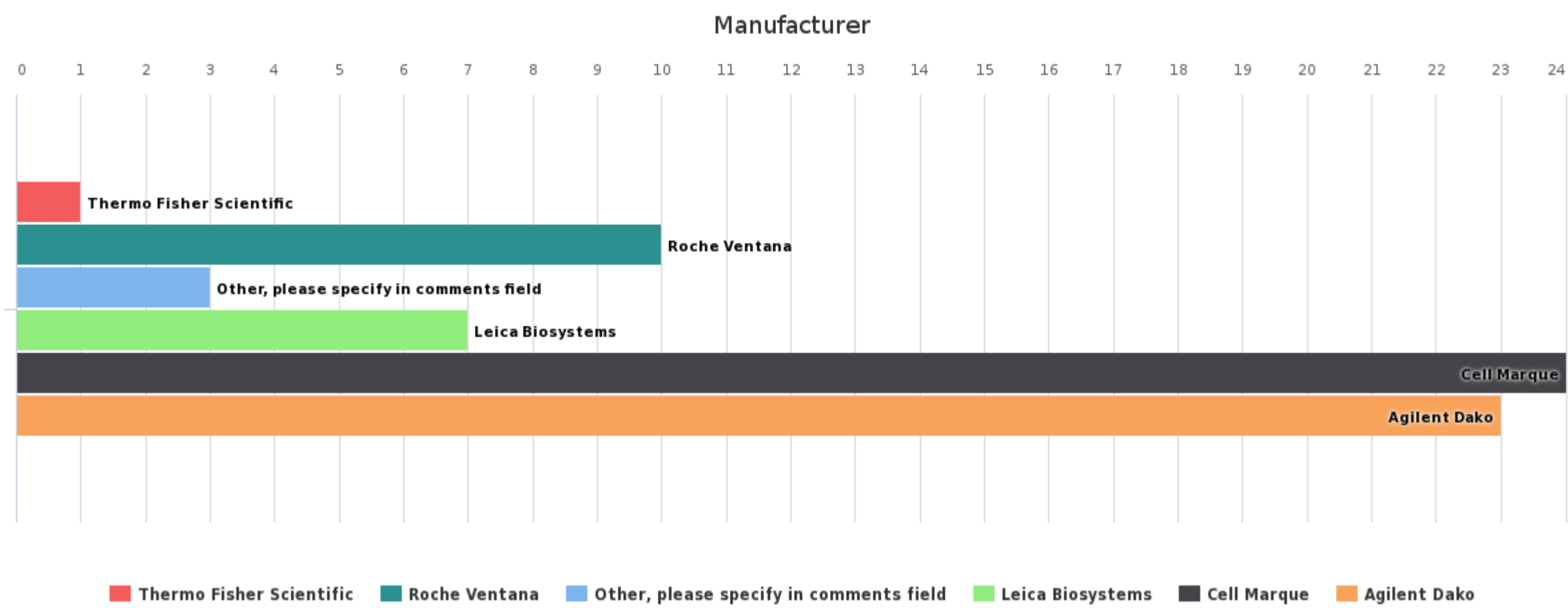


GENERAL DETAILS OF STAINING

Performance	Performance count
Automatic	63
Manual	6
Total:	69

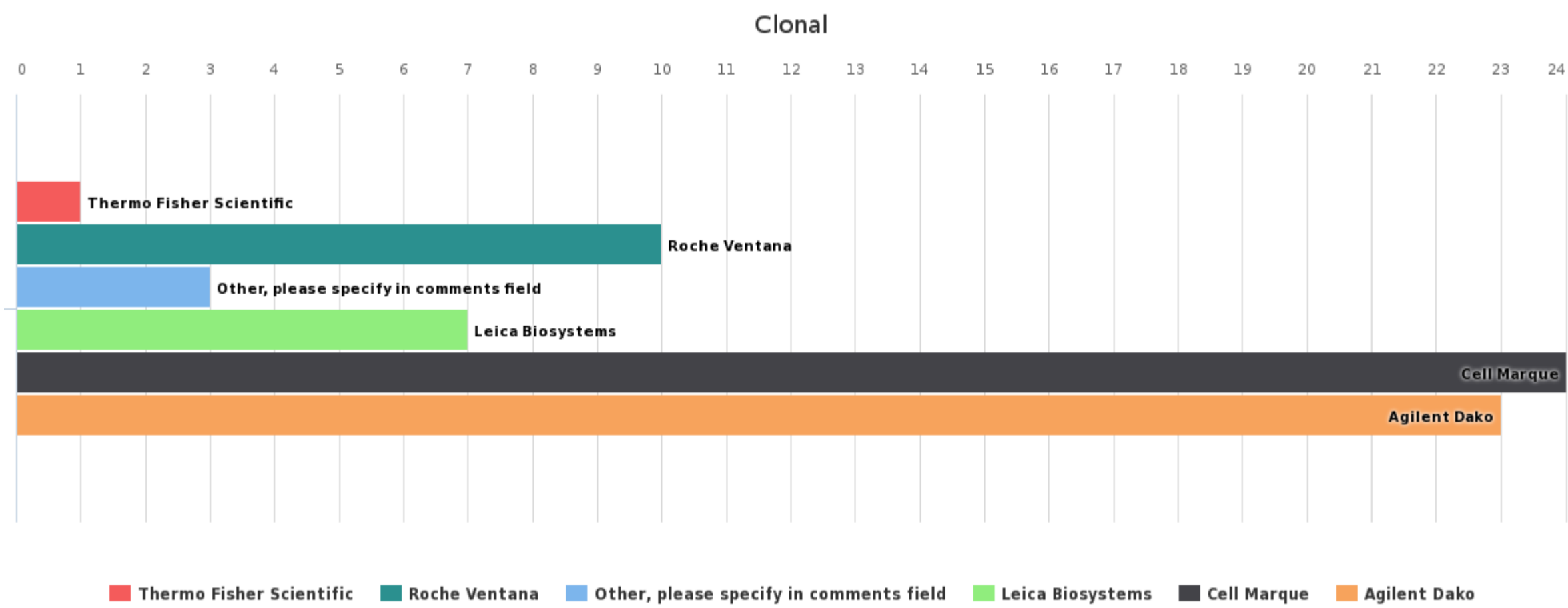


Instrument	Instrument count
Agilent Dako Autostainer Link 48	20
Agilent Dako Autostainer Plus	1
Agilent Dako Omnis	1
Leica BioSystems Bond III	5
Leica Biosystems Bondmax	3
Roche Ventana Benchmark Ultra	27
Roche Ventana Benchmark XT	6
Thermo Fisher Scientific Lab Vision Autostainer 480S-2D	1
Total:	64

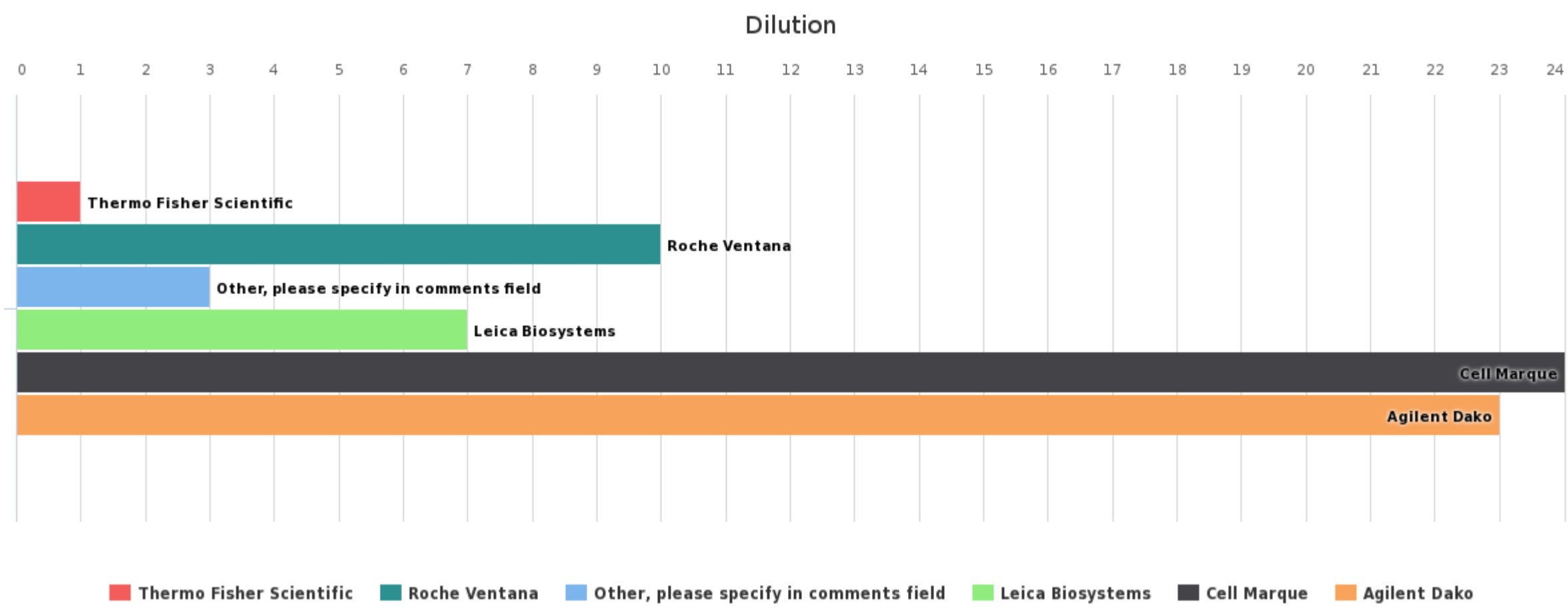


PRIMARY ANTIBODY

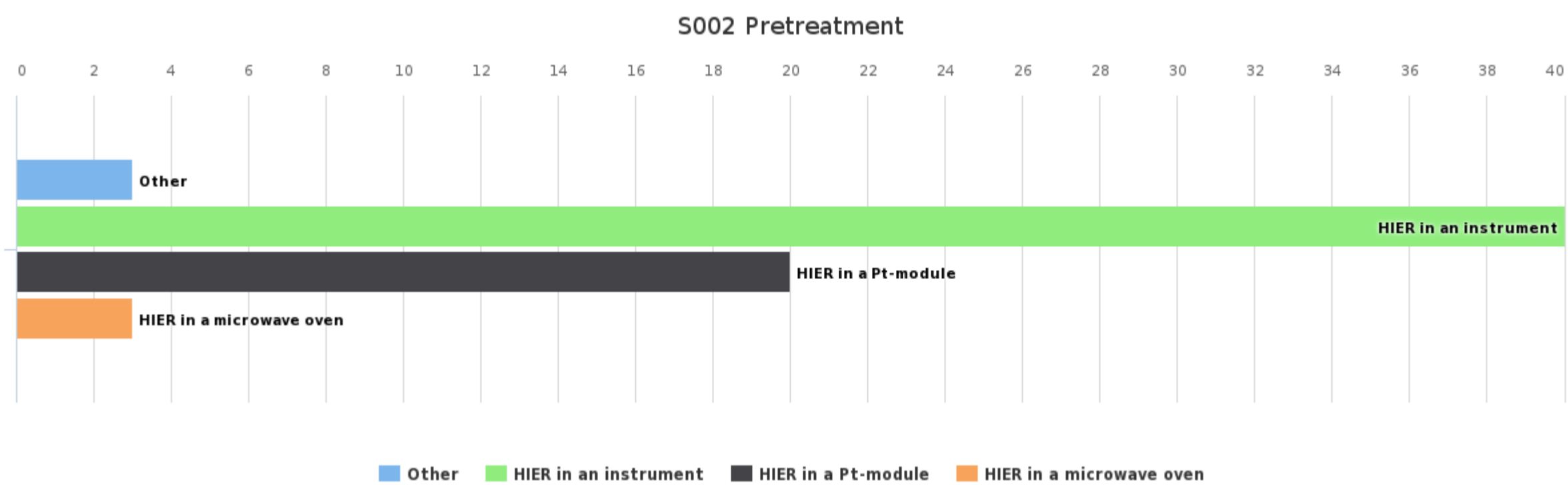
Manufacturer	Manufacturer count
Agilent Dako	23
Cell Marque	24
Leica Biosystems	7
Other, please specify in comments field	3
Roche Ventana	10
Thermo Fisher Scientific	1
Total:	68



Clonal	Clonal count
Monoclonal	68
Total:	68

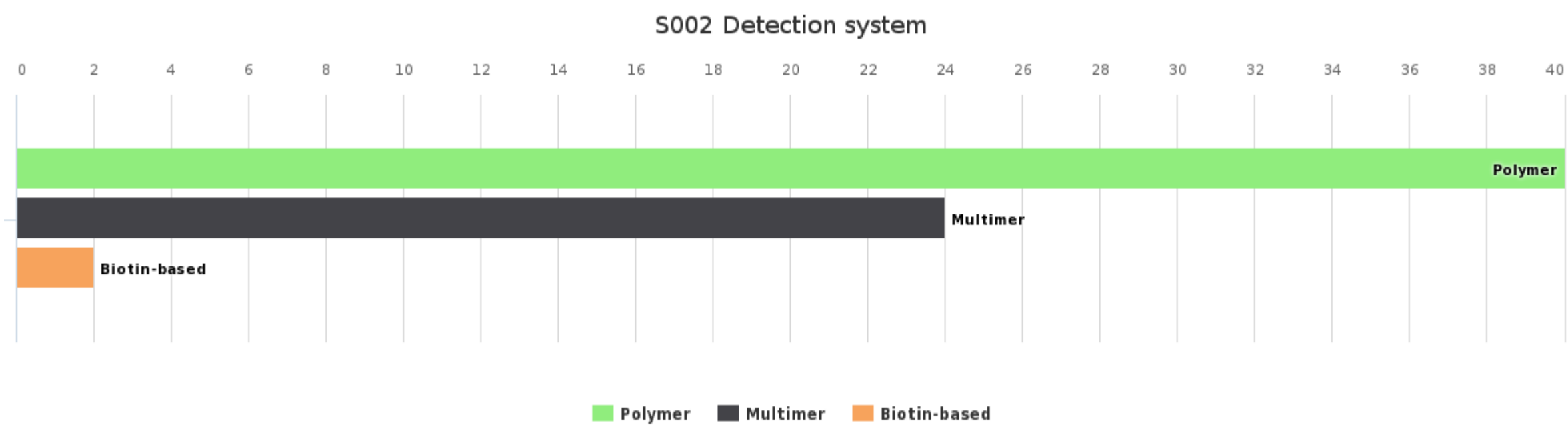


Dilution	Dilution count
Dilution	23
Ready-to-use	45
Total:	68



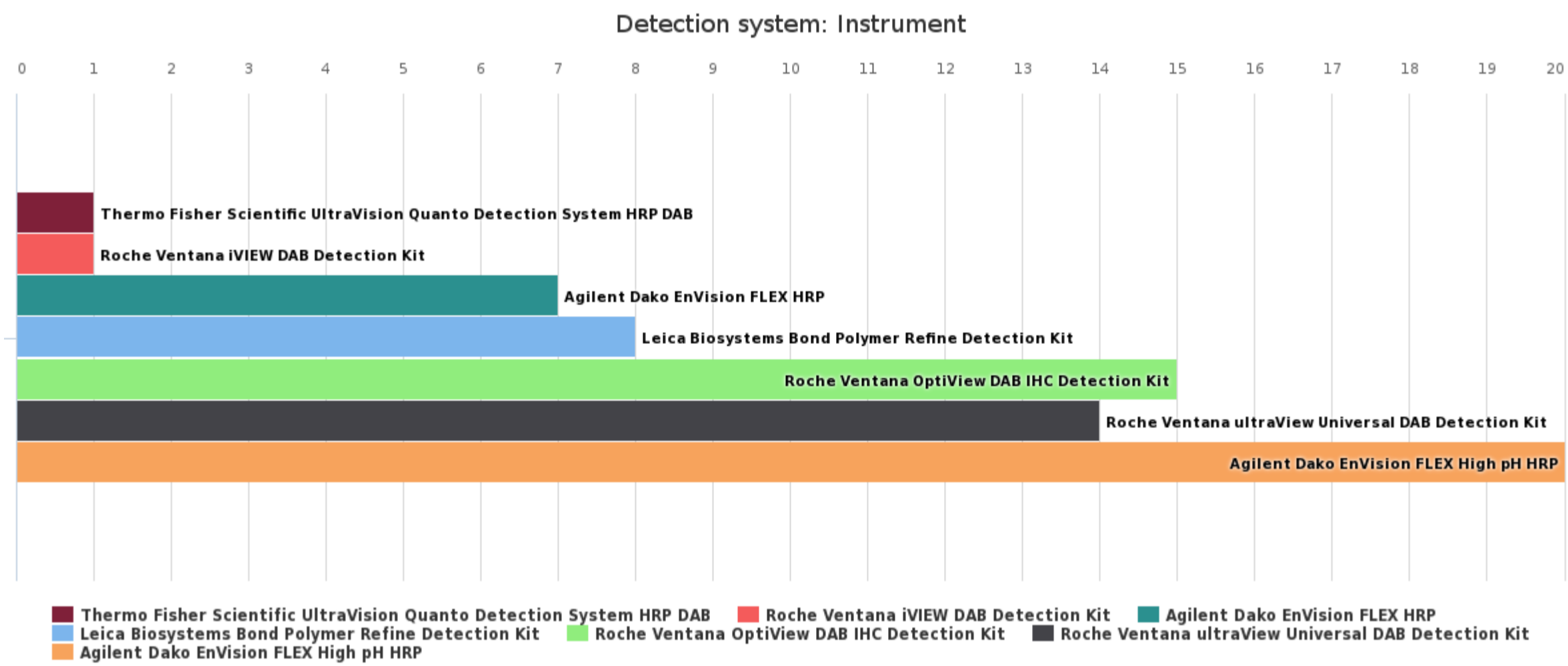
PRETREATMENT

Pretreatment	Pretreatment count
HIER in a microwave oven	3
HIER in a Pt-module	20
HIER in an instrument	40
Other	3
Total:	66

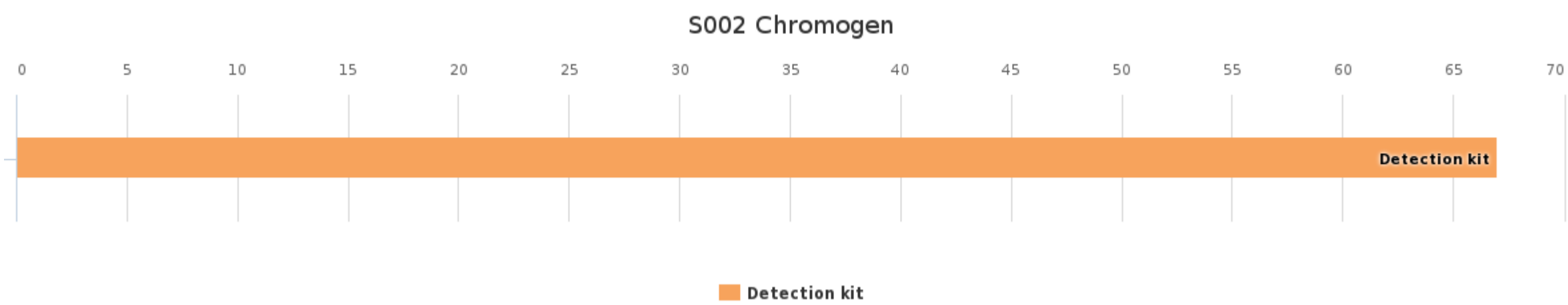


DETECTION SYSTEM

Detection system	Detection system count
Biotin-based	2
Multimer	24
Polymer	40
Total:	66

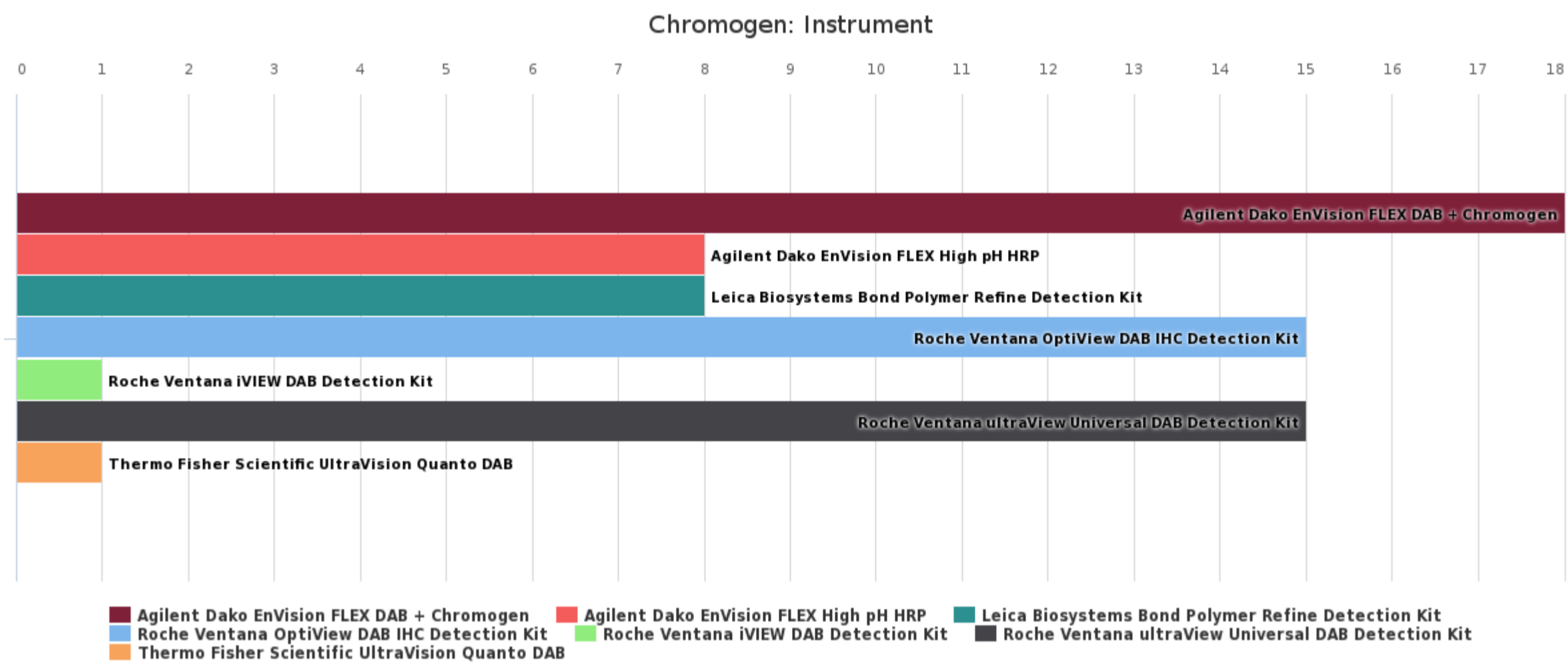


Detection system: Instrument	Detection system: Instrument count
Agilent Dako EnVision FLEX HRP	7
Agilent Dako EnVision FLEX High pH HRP	20
Leica Biosystems Bond Polymer Refine Detection Kit	8
Roche Ventana OptiView DAB IHC Detection Kit	15
Roche Ventana iVIEW DAB Detection Kit	1
Roche Ventana ultraView Universal DAB Detection Kit	14
Thermo Fisher Scientific UltraVision Quanto Detection System HRP DAB	1
Total:	66

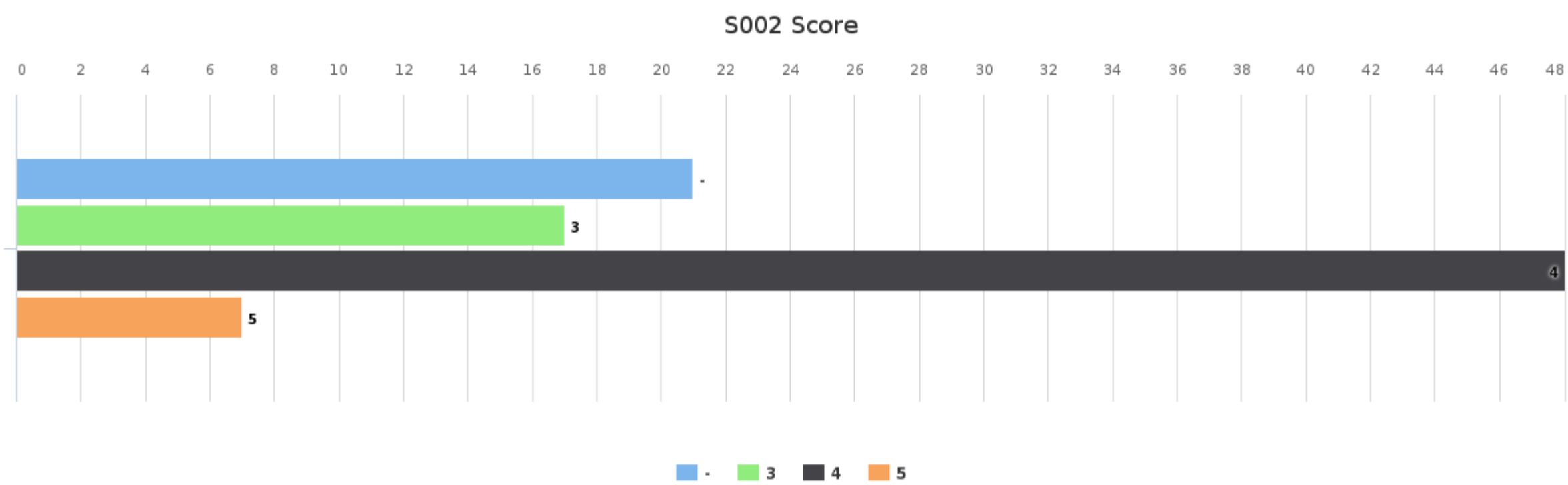


CHROMOGEN

Chromogen	Chromogen count
Detection kit	67
Total:	67

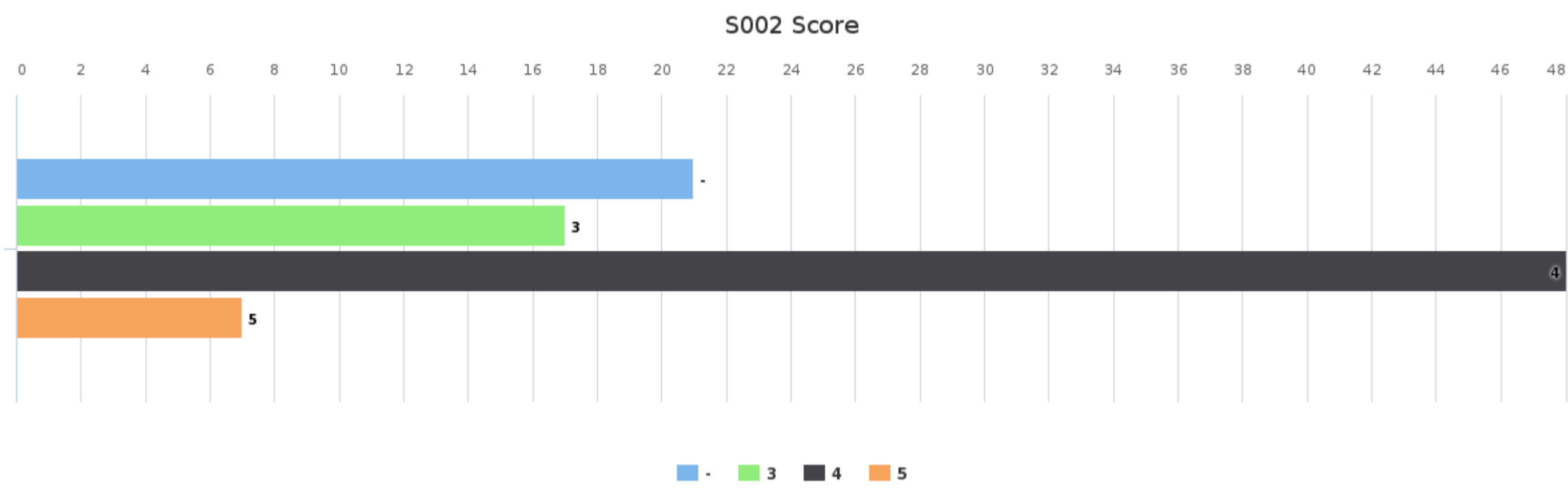


Chromogen: Instrument	Chromogen: Instrument count
Agilent Dako EnVision FLEX DAB + Chromogen	18
Agilent Dako EnVision FLEX High pH HRP	8
Leica Biosystems Bond Polymer Refine Detection Kit	8
Roche Ventana OptiView DAB IHC Detection Kit	15
Roche Ventana iVIEW DAB Detection Kit	1
Roche Ventana ultraView Universal DAB Detection Kit	15
Thermo Fisher Scientific UltraVision Quanto DAB	1
Total:	66



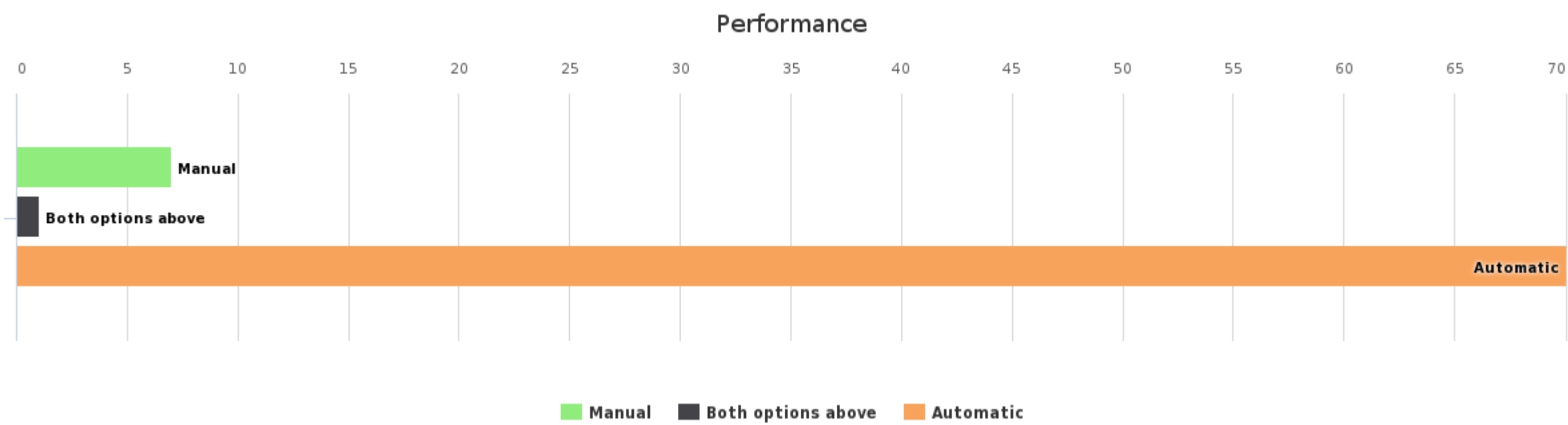
SCORE

Score	Comment
-	
	not available
3	
	background staining in kidney sample
	background staining in kidney sample, overstained
	contaminated from some other sample?, some background staining, stain deposits
	overstained
	some background staining in kidney sample, overstained
	some background staining in kidney sample, slightly overstained
	some background staining in kidney sample, understained, dark hematoxylin
	some background staining in kidney sample, uneven
	some background staining, uneven
	understained
	understained, dark hematoxylin, some background staining in kidney sample
	understained, false positive staining in fallobian tube
	understained, uneven
	undertaining
4	
	background staining in kidney sample
	hematoxylin is quite weak
	slightly overstained, some background staining in kidney sample
	slightly overstained, uneven
	slightly understained
	slightly understained, some background staining in kidney sample
	slightly understained, uneven
	slightly uneven
	sligtly understained
	some background in kidney sample, slightly understained
	some background in kidney sample, sligthly understained
	some background staining
	some background staining in kidney sample
	some background staining in kidney sample, follobian tube sample is uneven
	some background staining in kidney sample, slightly overstained
	some background staining in kidney sample, slightly understained
	some background staining in kidney sample, uneven staining in epithelial cells in fallobian tube
	some background staining, slightly eneven
	uneven staining in epithelial cells in fallobian tube
5	
	Excellent
Total:	



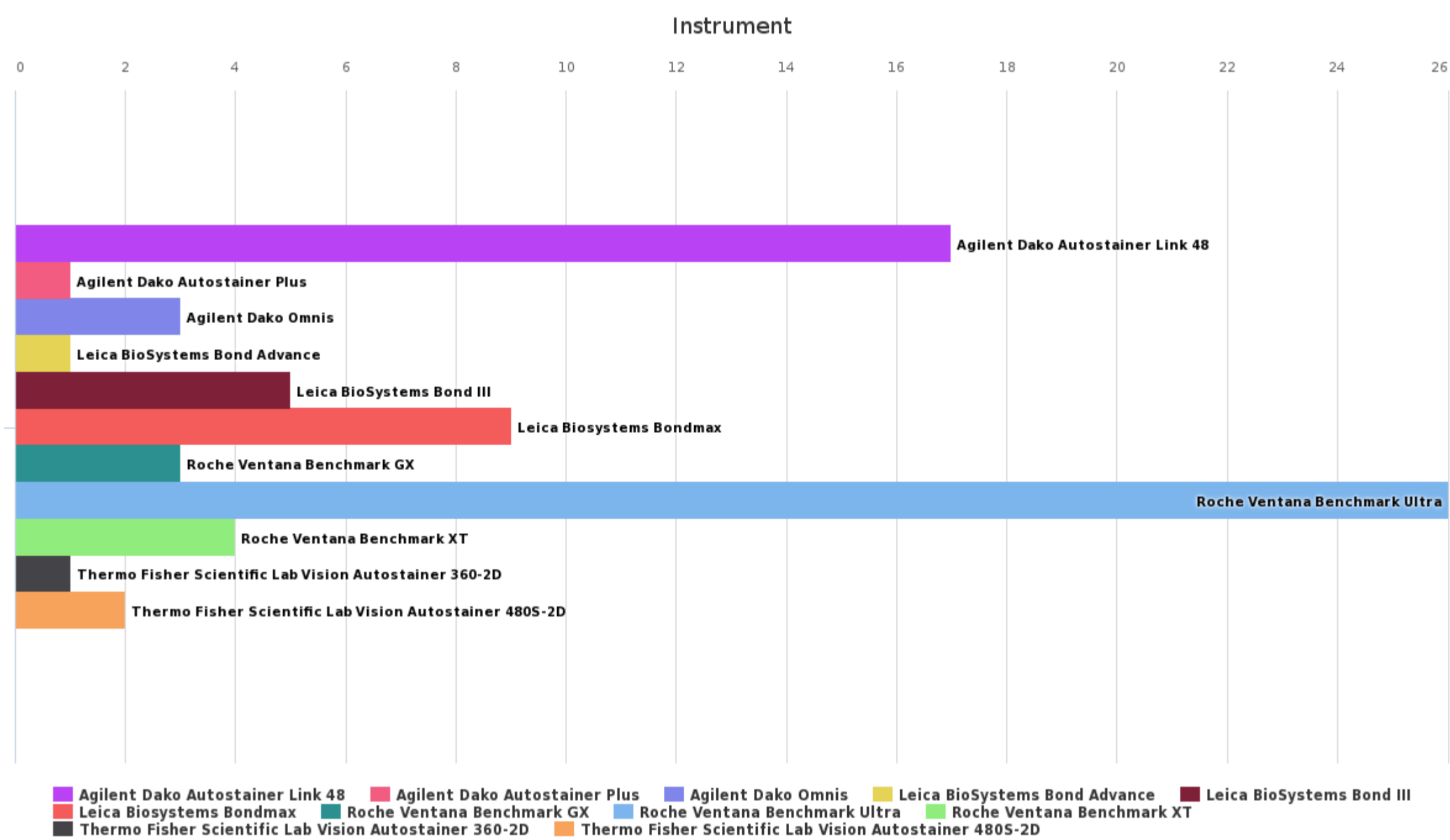
Score	Score count
-	21
3	17
4	48
5	7
Total:	93

S003 | Sample S003 CK20



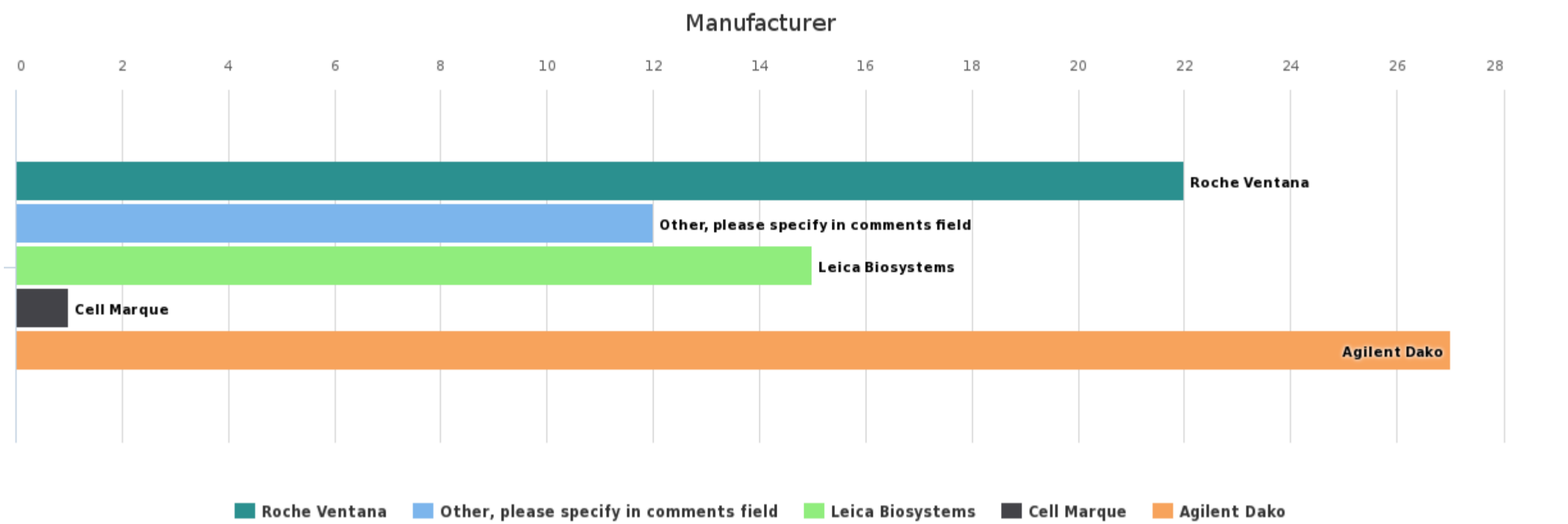
GENERAL DETAILS OF STAINING

Performance	Performance count
Automatic	70
Both options above	1
Manual	7
Total:	78



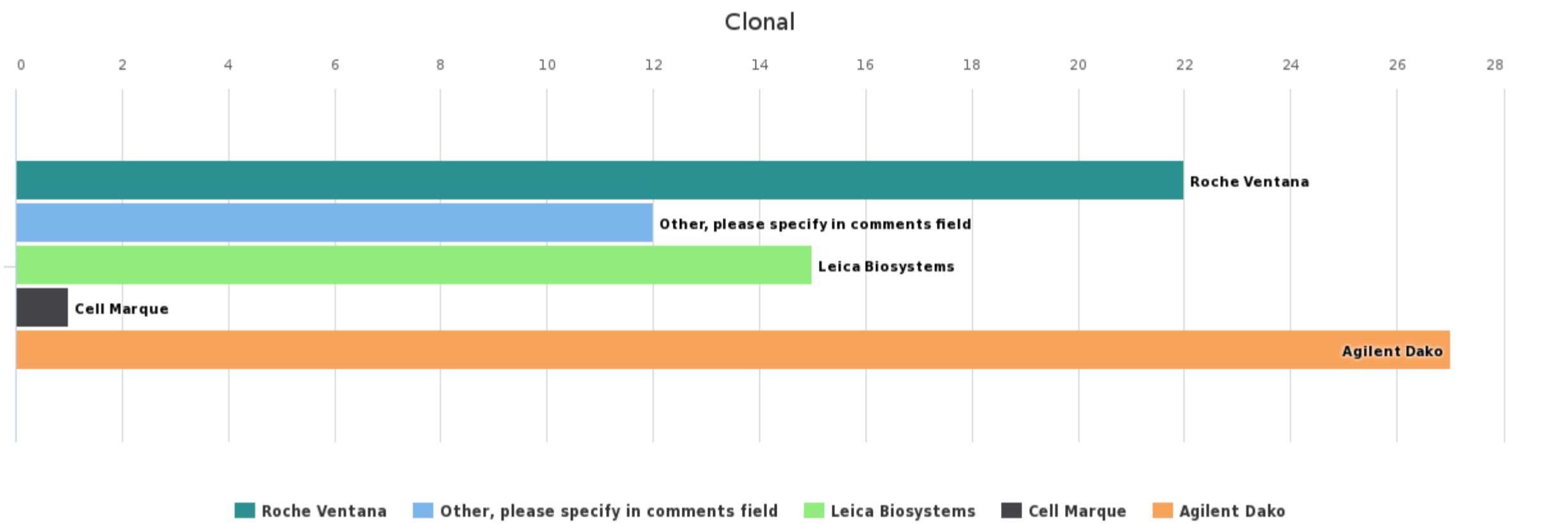
Instrument	Instrument count
Agilent Dako Autostainer Link 48	17
Agilent Dako Autostainer Plus	1
Agilent Dako Omnis	3
Leica BioSystems Bond Advance	1
Leica BioSystems Bond III	5
Leica Biosystems Bondmax	9
Roche Ventana Benchmark GX	3
Roche Ventana Benchmark Ultra	26

Roche Ventana Benchmark XT	4
Thermo Fisher Scientific Lab Vision Autostainer 360-2D	1
Thermo Fisher Scientific Lab Vision Autostainer 480S-2D	2
Total:	72

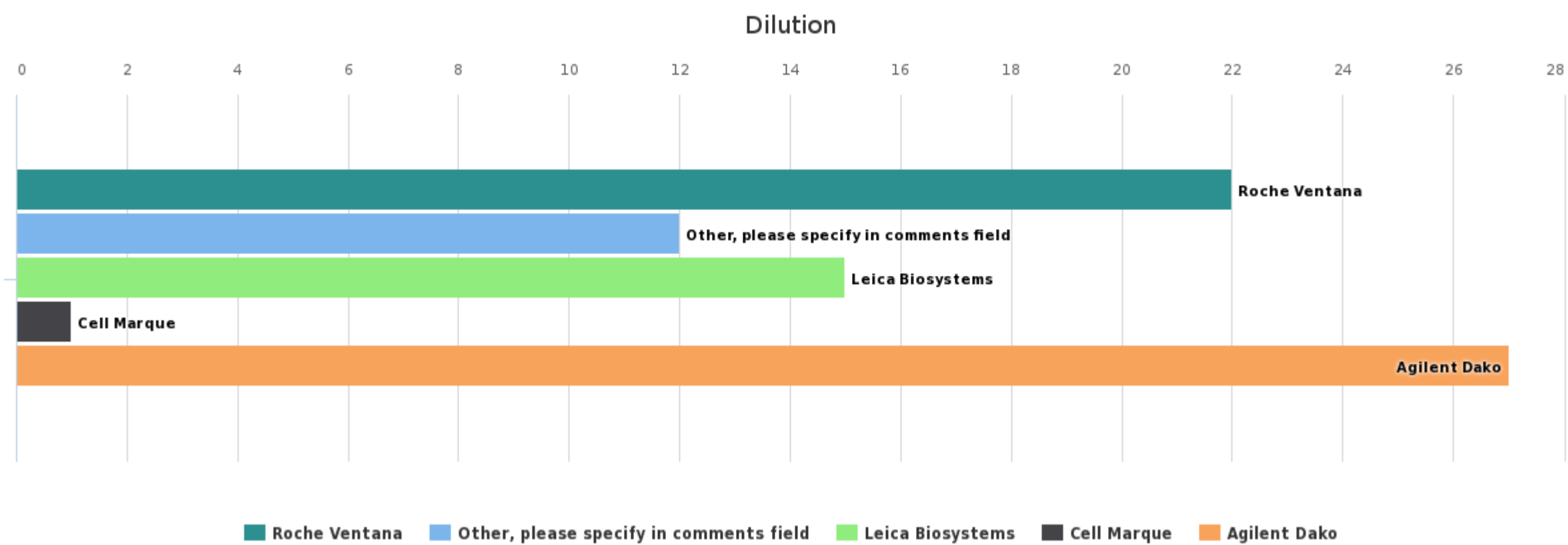


PRIMARY ANTIBODY

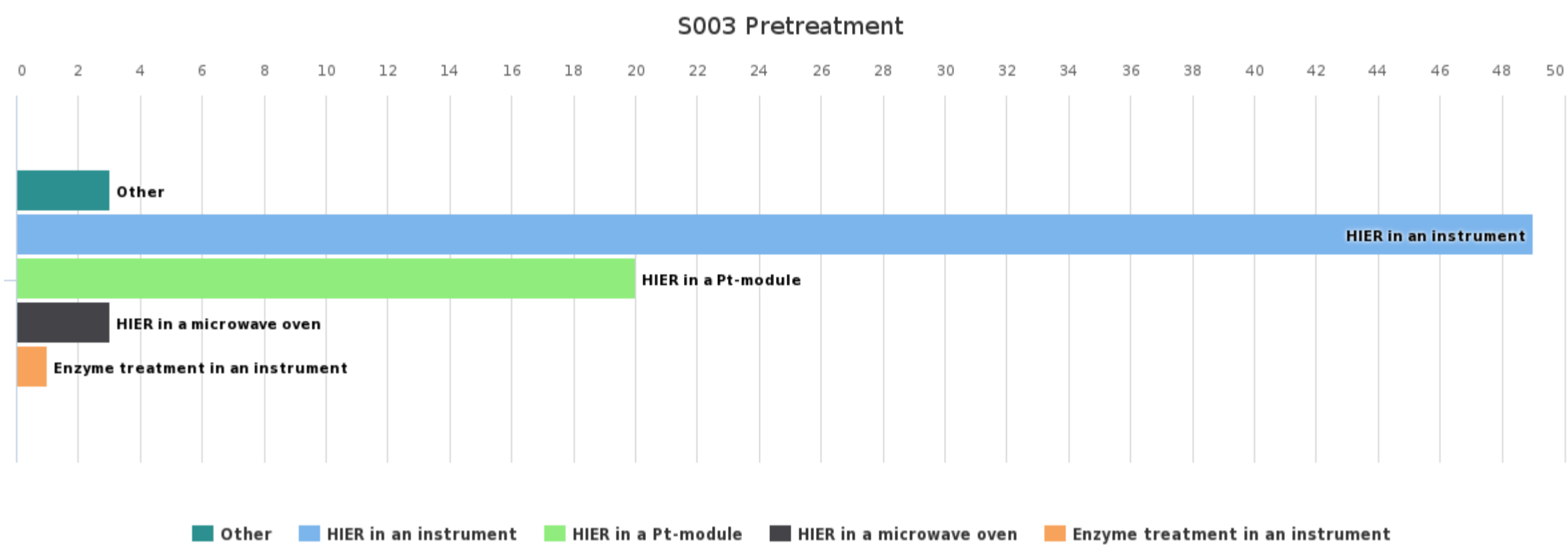
Manufacturer	Manufacturer count
Agilent Dako	27
Cell Marque	1
Leica Biosystems	15
Other, please specify in comments field	12
Roche Ventana	22
Total:	77



Clonal	Clonal count
Monoclonal	75
Polyclonal	1
Total:	76

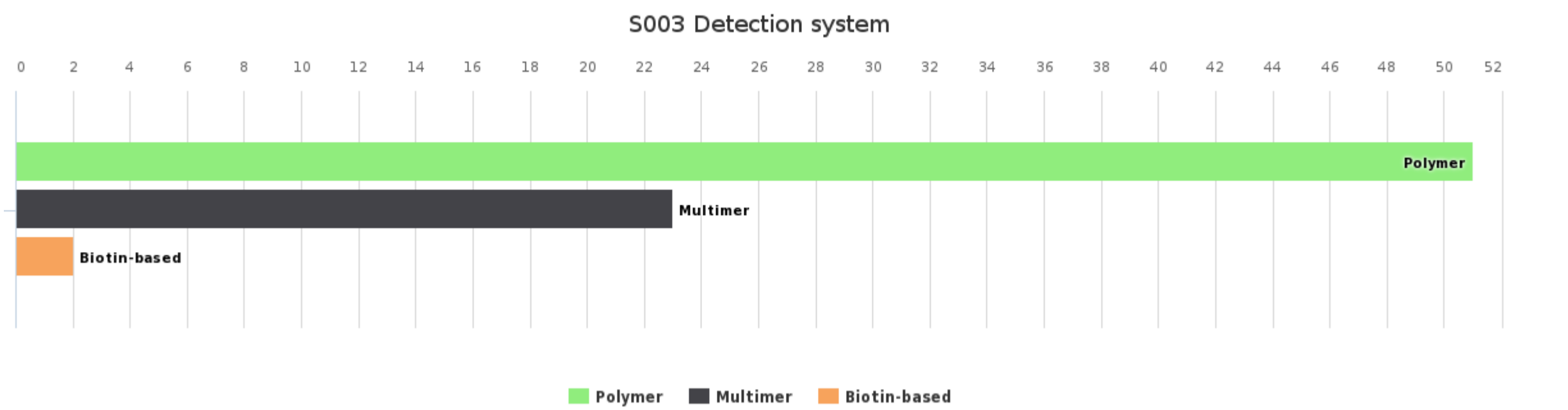


Dilution	Dilution count
Dilution	31
Ready-to-use	46
Total:	77



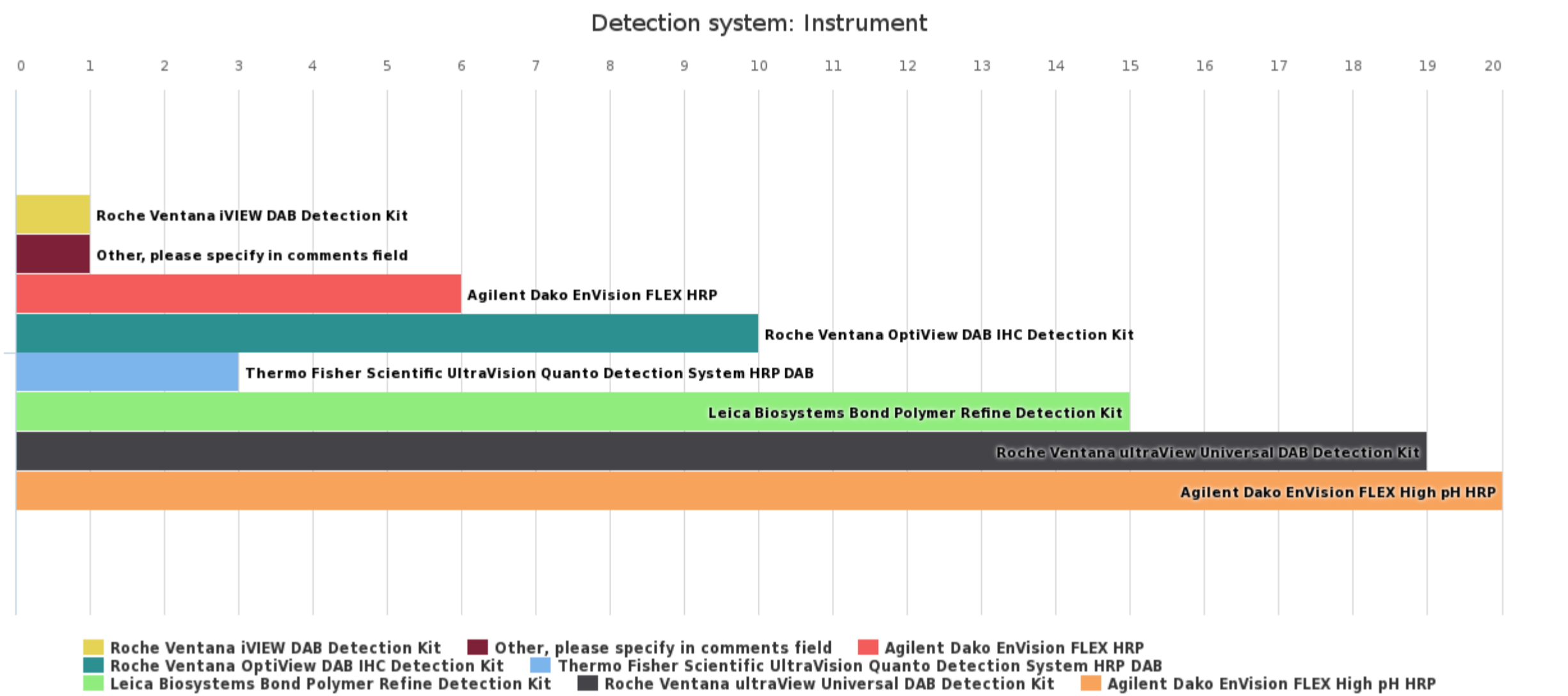
PRETREATMENT

Pretreatment	Pretreatment count
Enzyme treatment in an instrument	1
HIER in a microwave oven	3
HIER in a Pt-module	20
HIER in an instrument	49
Other	3
Total:	76

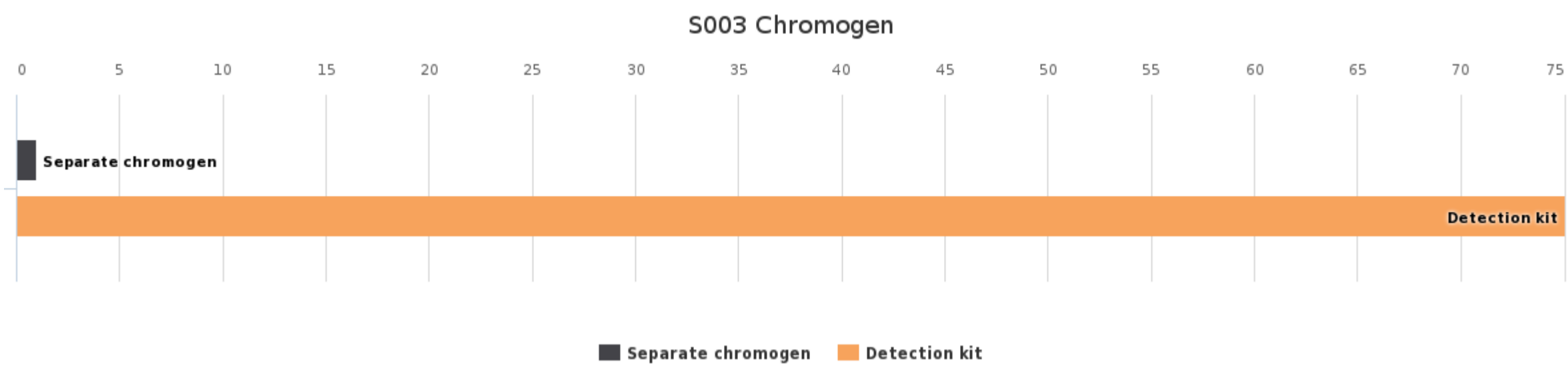


DETECTION SYSTEM

Detection system	Detection system count
Biotin-based	2
Multimer	23
Polymer	51
Total:	76

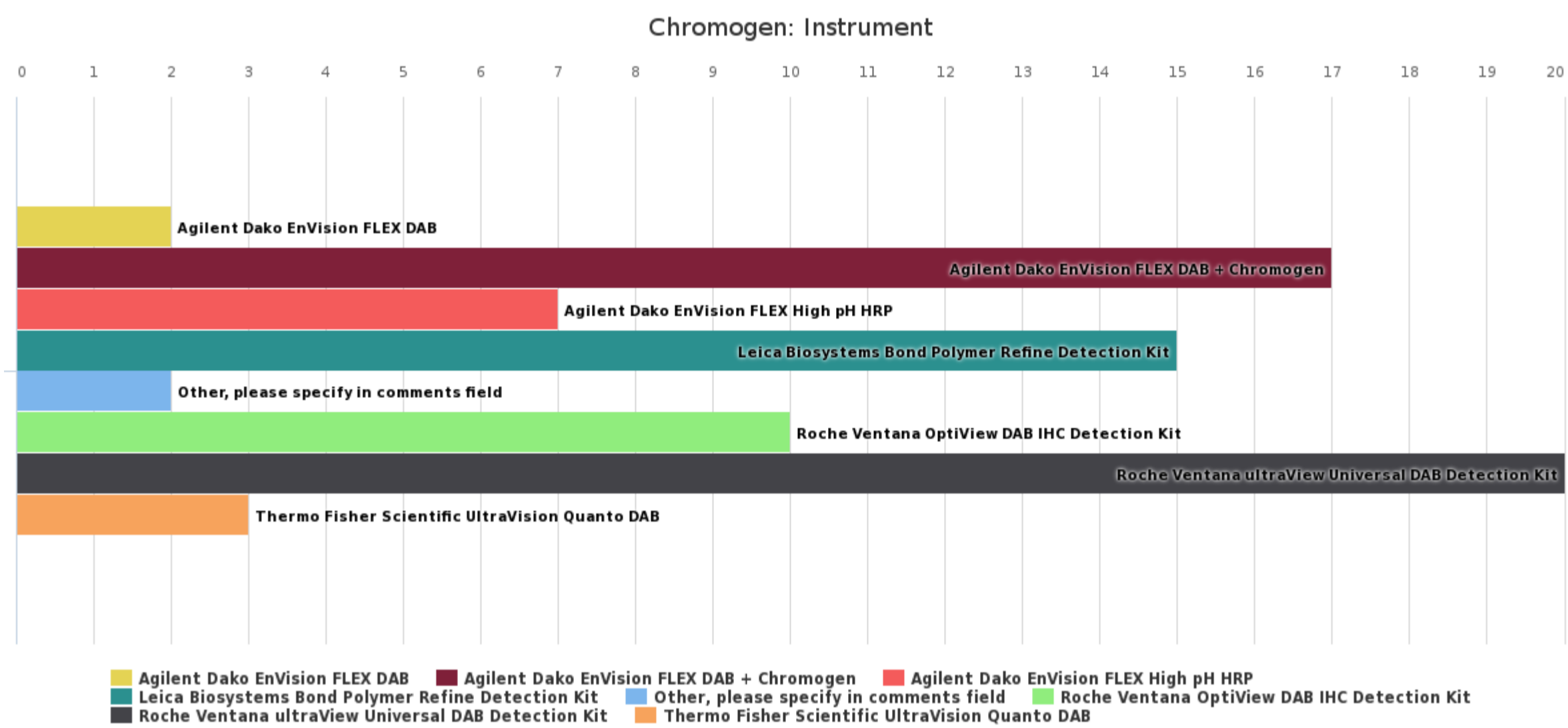


Detection system: Instrument	Detection system: Instrument count
Agilent Dako EnVision FLEX HRP	6
Agilent Dako EnVision FLEX High pH HRP	20
Leica Biosystems Bond Polymer Refine Detection Kit	15
Other, please specify in comments field	1
Roche Ventana OptiView DAB IHC Detection Kit	10
Roche Ventana iVIEW DAB Detection Kit	1
Roche Ventana ultraView Universal DAB Detection Kit	19
Thermo Fisher Scientific UltraVision Quanto Detection System HRP DAB	3
Total:	75

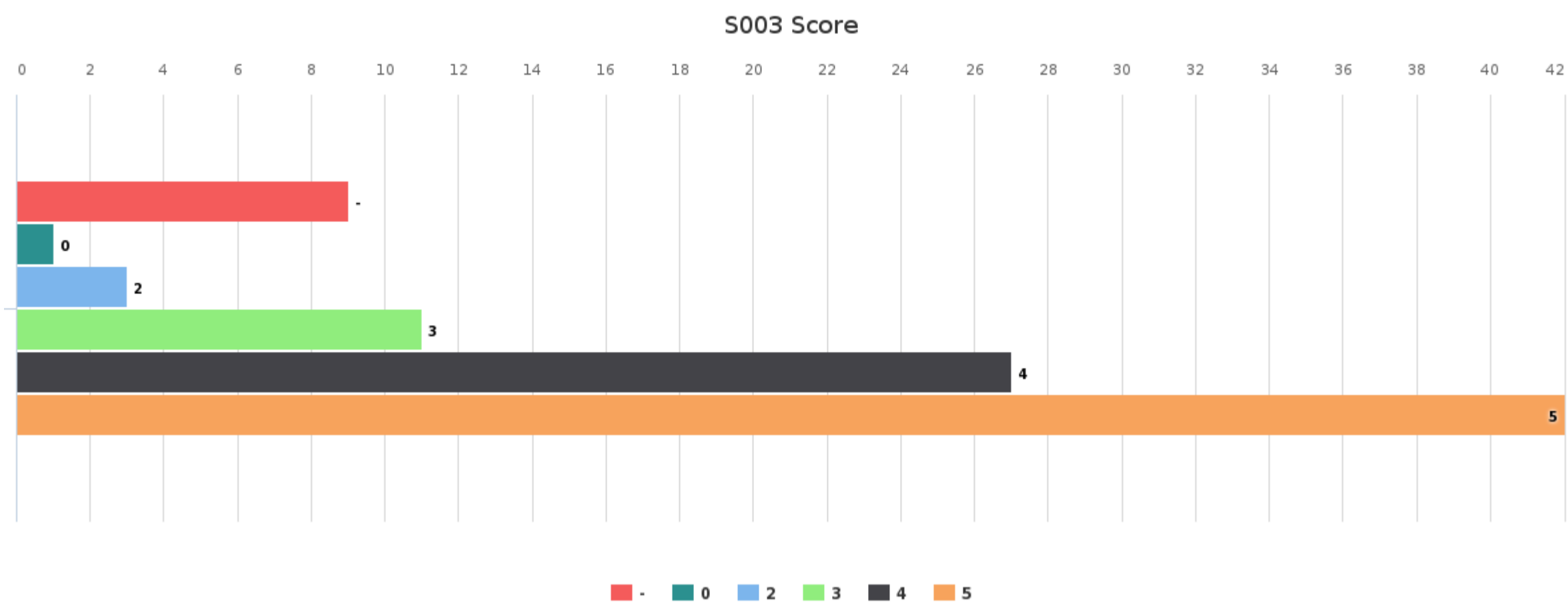


CHROMOGEN

Chromogen	Chromogen count
Detection kit	75
Separate chromogen	1
Total:	76

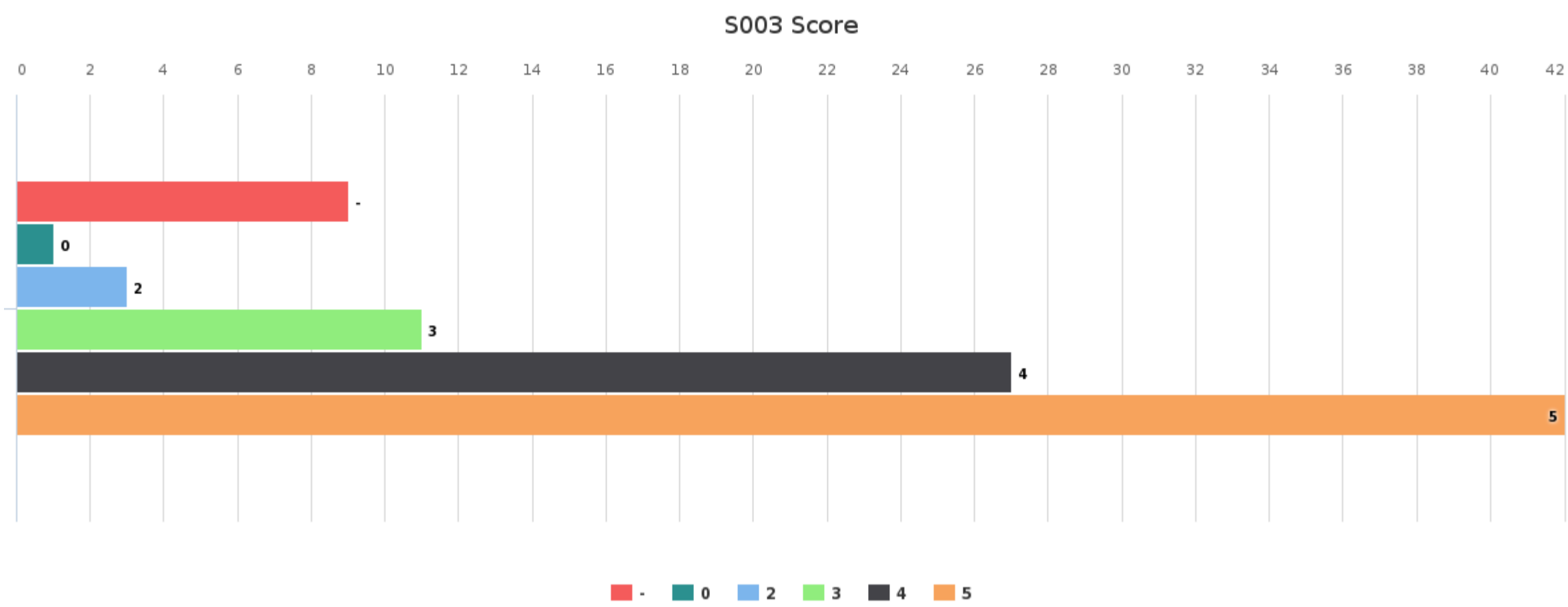


Chromogen: Instrument	Chromogen: Instrument count
Agilent Dako EnVision FLEX DAB	2
Agilent Dako EnVision FLEX DAB + Chromogen	17
Agilent Dako EnVision FLEX High pH HRP	7
Leica Biosystems Bond Polymer Refine Detection Kit	15
Other, please specify in comments field	2
Roche Ventana OptiView DAB IHC Detection Kit	10
Roche Ventana ultraView Universal DAB Detection Kit	20
Thermo Fisher Scientific UltraVision Quanto DAB	3
Total:	76



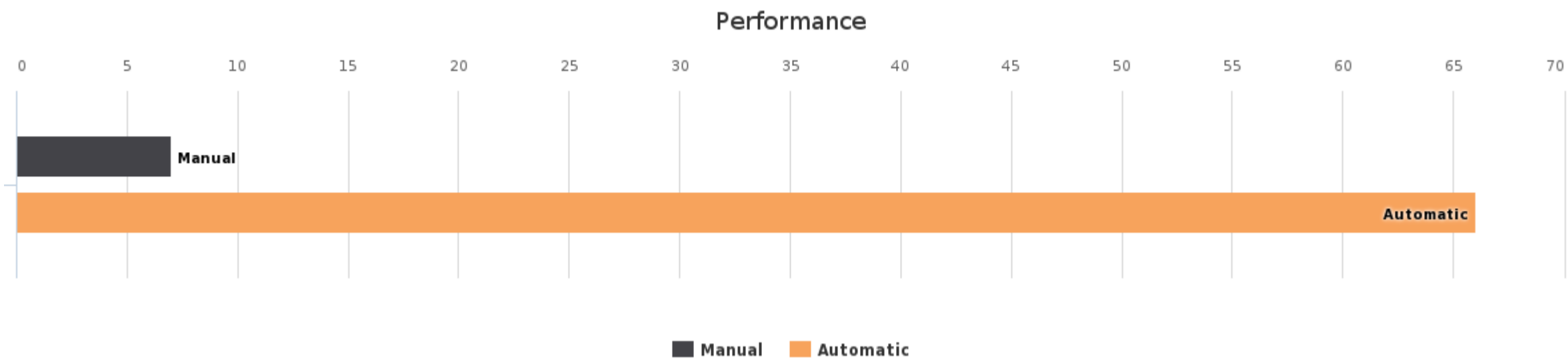
SCORE

Score	Comment
-	
	not available
0	
	negative
2	
	false positivity in muscle cells
	understained, uneven, background staining
	weak staining
3	
	slightly understained, stain deposits
	understained
	understained, dark hematoxylin
	understained, slightly dark hematoxylin
	understaining
	undertained
4	
	slightly understained
	slightly understained, dark hematoxylin
	slightly understained, some background staining
	weak hematoxylin in tumor sample
5	
	Excellent
	hematoxylin is quite weak
Total:	



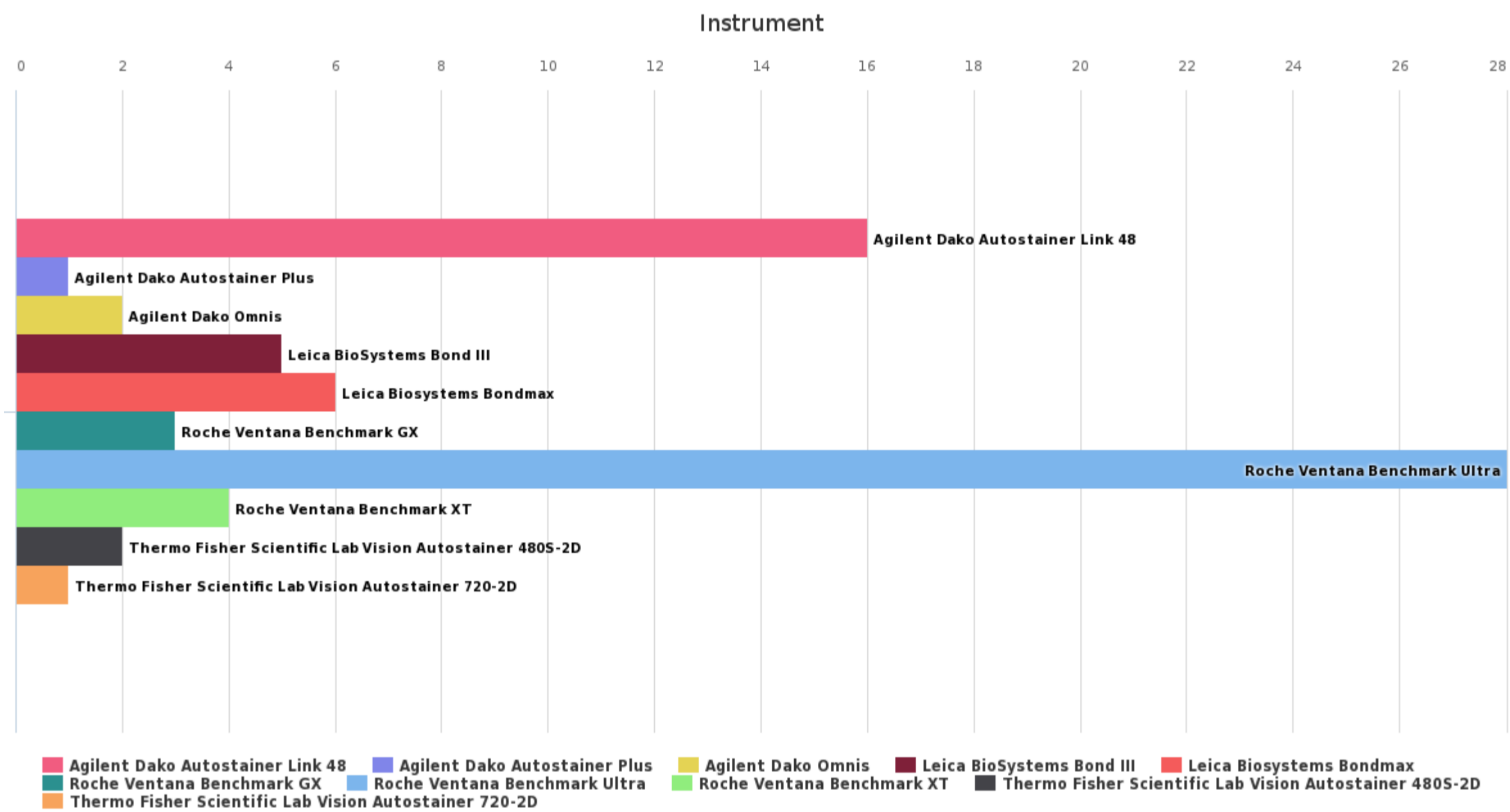
Score	Score count
-	9
0	1
2	3
3	11
4	27
5	42
Total:	93

S004 | Sample S004 CDX2

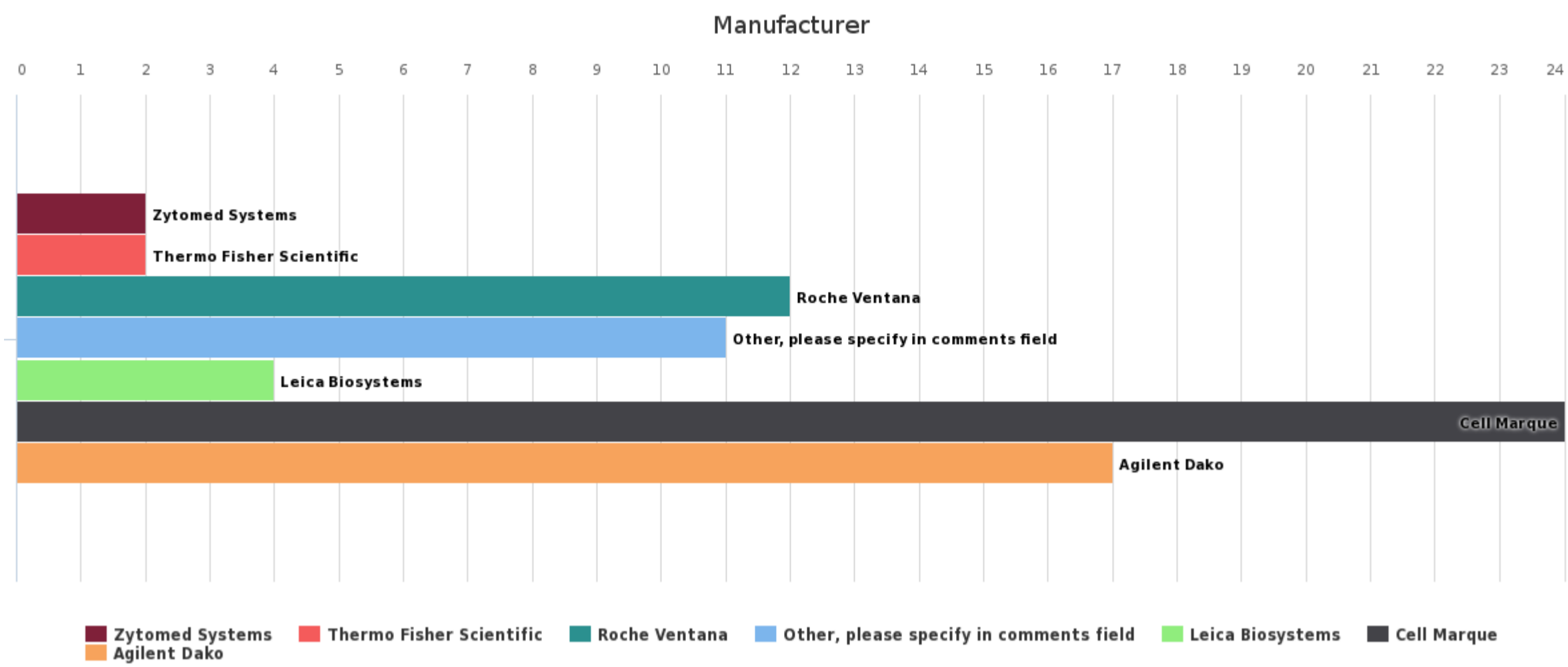


GENERAL DETAILS OF STAINING

Performance	Performance count
Automatic	66
Manual	7
Total:	73

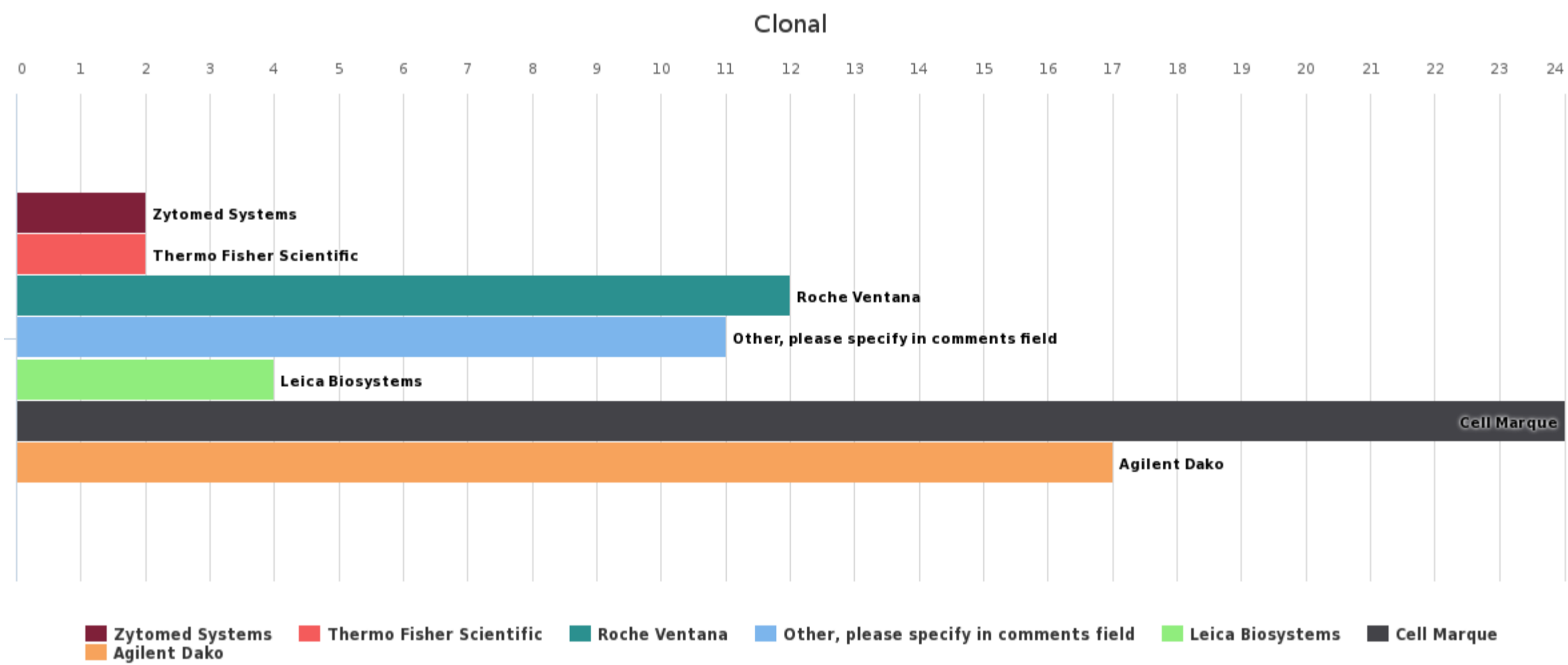


Instrument	Instrument count
Agilent Dako Autostainer Link 48	16
Agilent Dako Autostainer Plus	1
Agilent Dako Omnis	2
Leica BioSystems Bond III	5
Leica Biosystems Bondmax	6
Roche Ventana Benchmark GX	3
Roche Ventana Benchmark Ultra	28
Roche Ventana Benchmark XT	4
Thermo Fisher Scientific Lab Vision Autostainer 480S-2D	2
Thermo Fisher Scientific Lab Vision Autostainer 720-2D	1
Total:	68

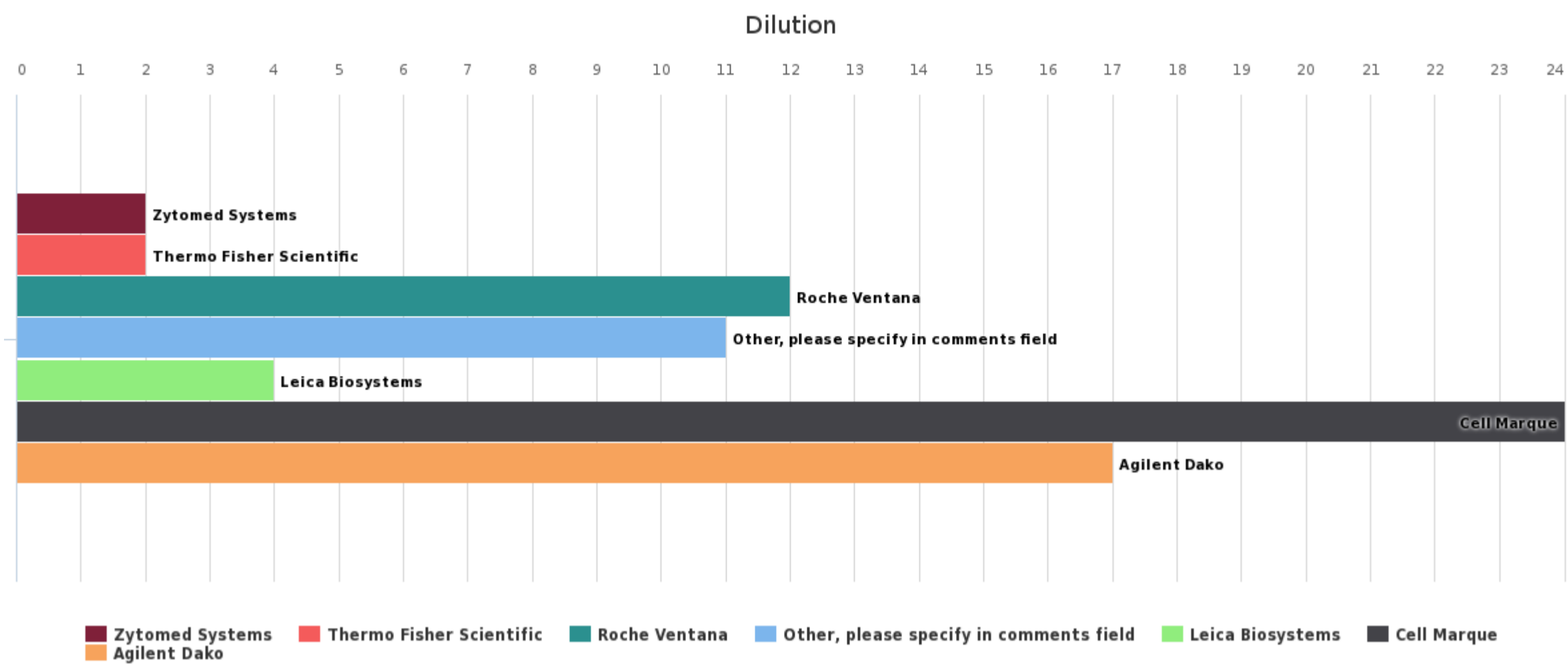


PRIMARY ANTIBODY

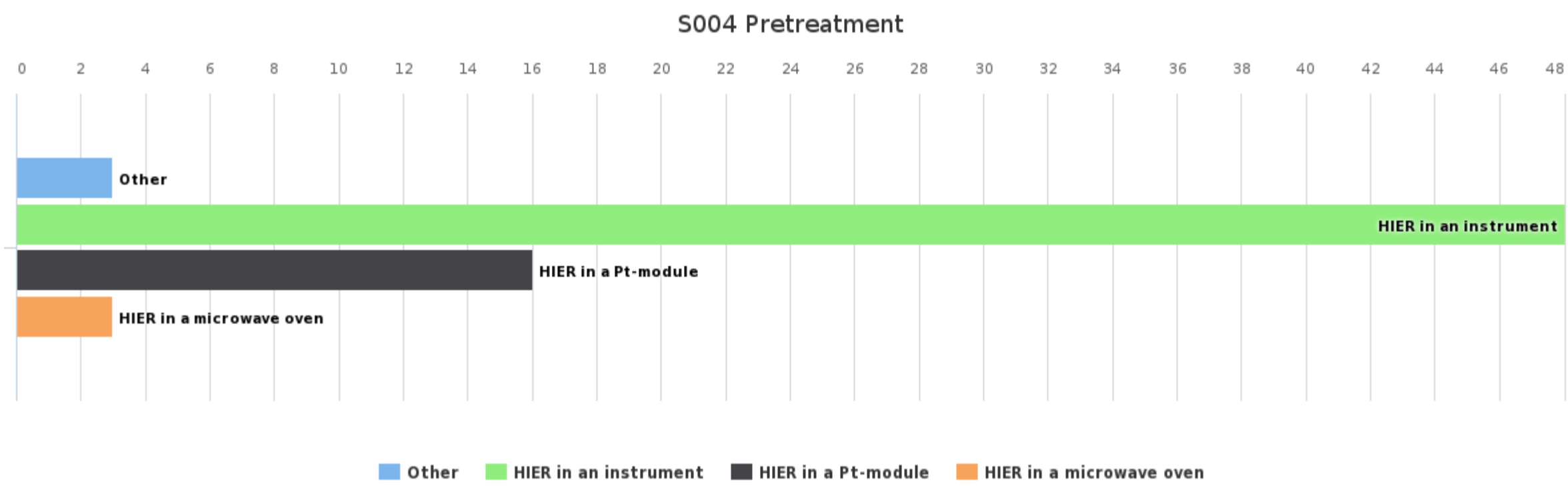
Manufacturer	Manufacturer count
Agilent Dako	17
Cell Marque	24
Leica Biosystems	4
Other, please specify in comments field	11
Roche Ventana	12
Thermo Fisher Scientific	2
Zytomed Systems	2
Total:	72



Clonal	Clonal count
Monoclonal	70
Polyclonal	1
Total:	71

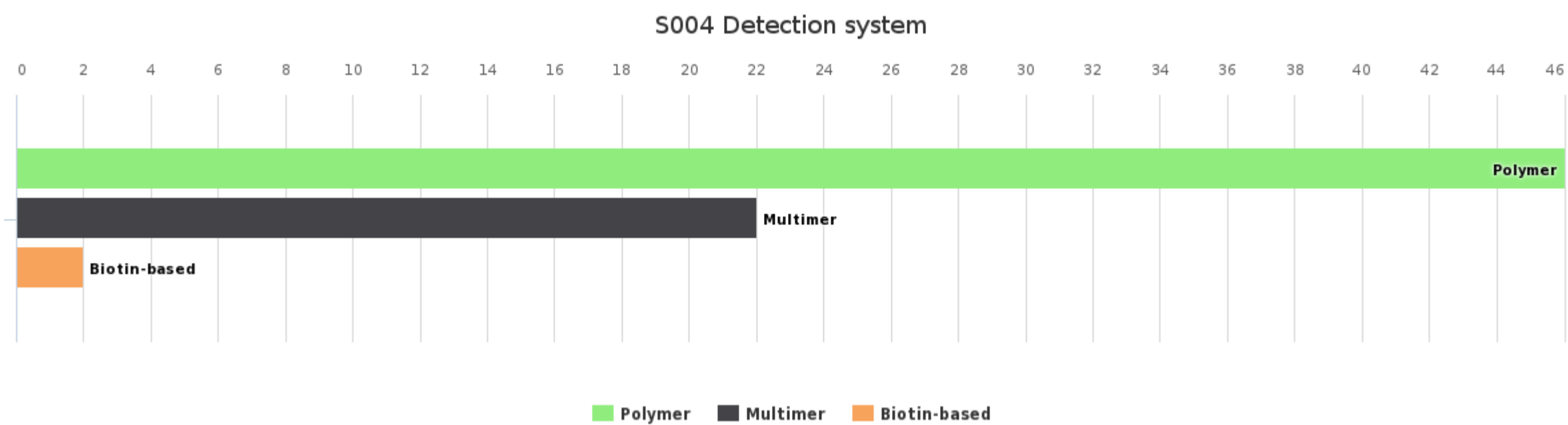


Dilution	Dilution count
Dilution	28
Ready-to-use	44
Total:	72



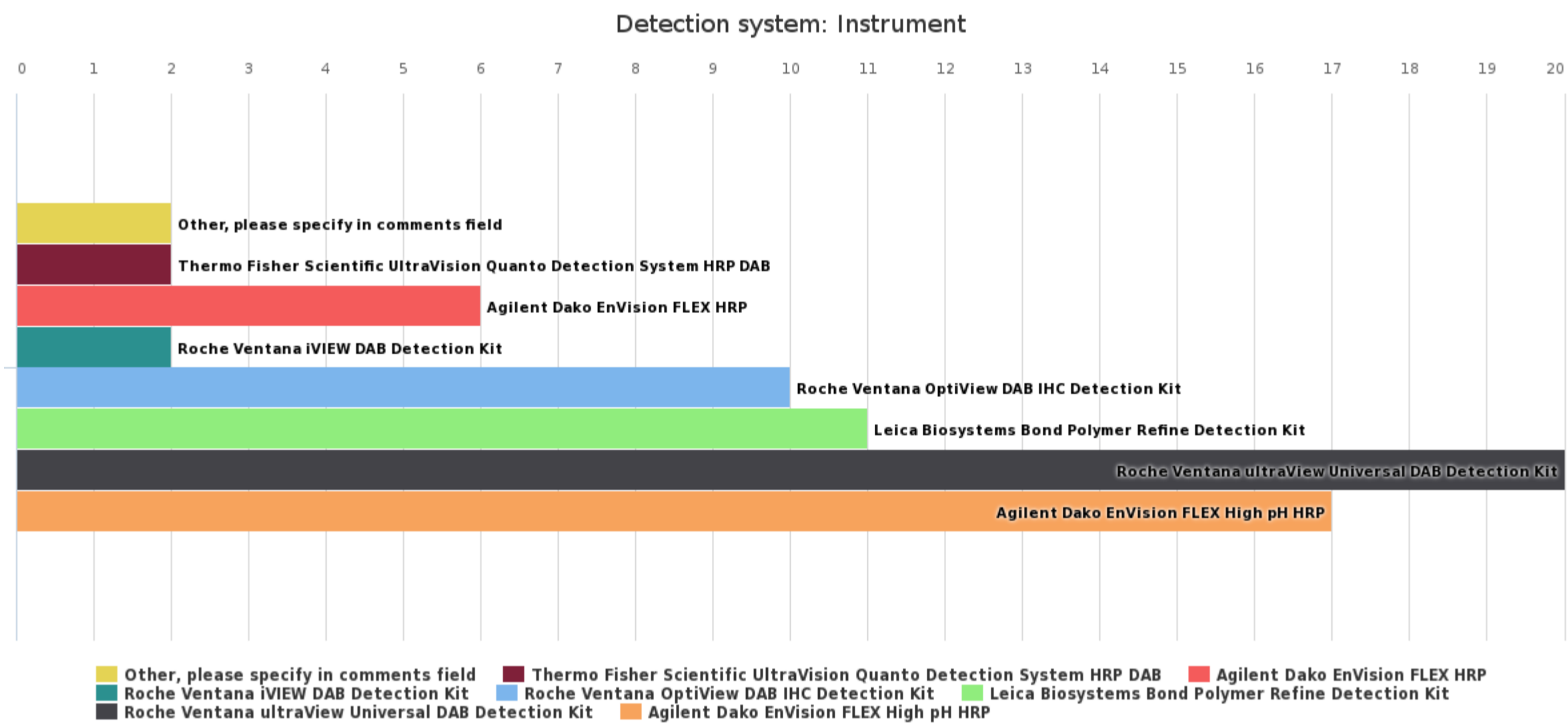
PRETREATMENT

Pretreatment	Pretreatment count
HIER in a microwave oven	3
HIER in a Pt-module	16
HIER in an instrument	48
Other	3
Total:	70

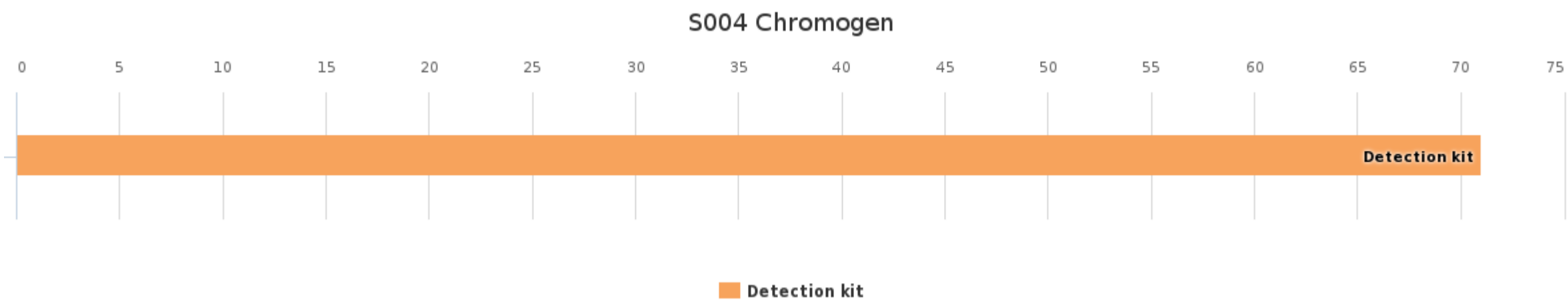


DETECTION SYSTEM

Detection system	Detection system count
Biotin-based	2
Multimer	22
Polymer	46
Total:	70

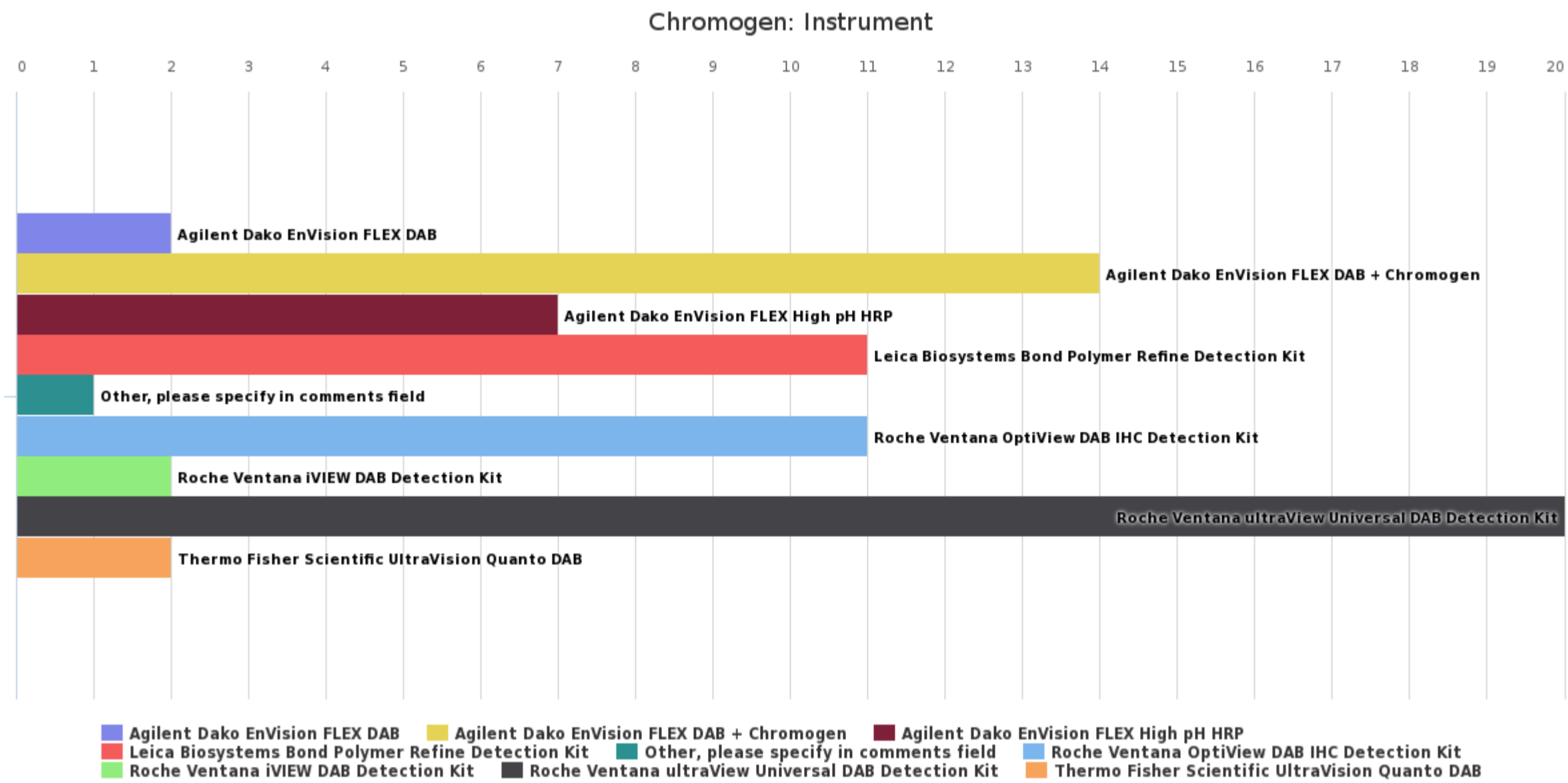


Detection system: Instrument	Detection system: Instrument count
Agilent Dako EnVision FLEX HRP	6
Agilent Dako EnVision FLEX High pH HRP	17
Leica Biosystems Bond Polymer Refine Detection Kit	11
Other, please specify in comments field	2
Roche Ventana OptiView DAB IHC Detection Kit	10
Roche Ventana iVIEW DAB Detection Kit	2
Roche Ventana ultraView Universal DAB Detection Kit	20
Thermo Fisher Scientific UltraVision Quanto Detection System HRP DAB	2
Total:	70

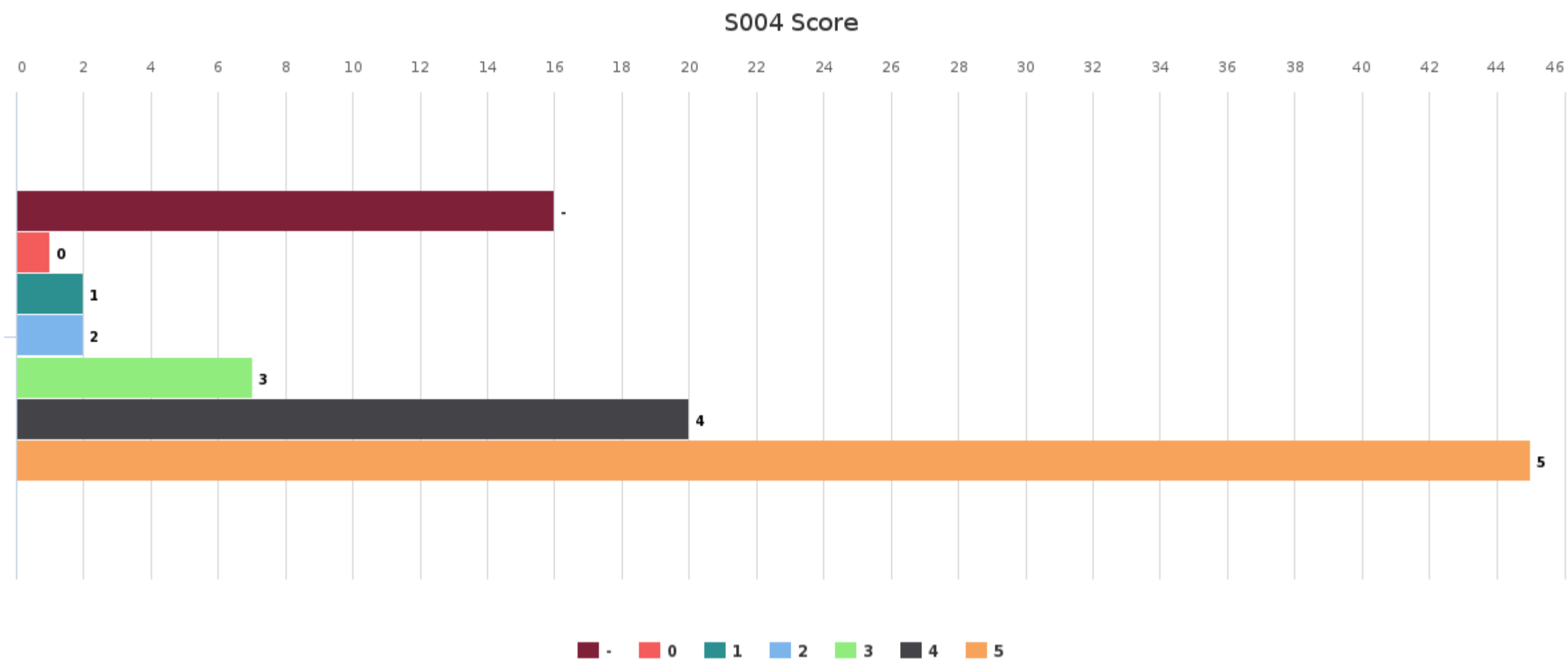


CHROMOGEN

Chromogen	Chromogen count
Detection kit	71
Total:	71

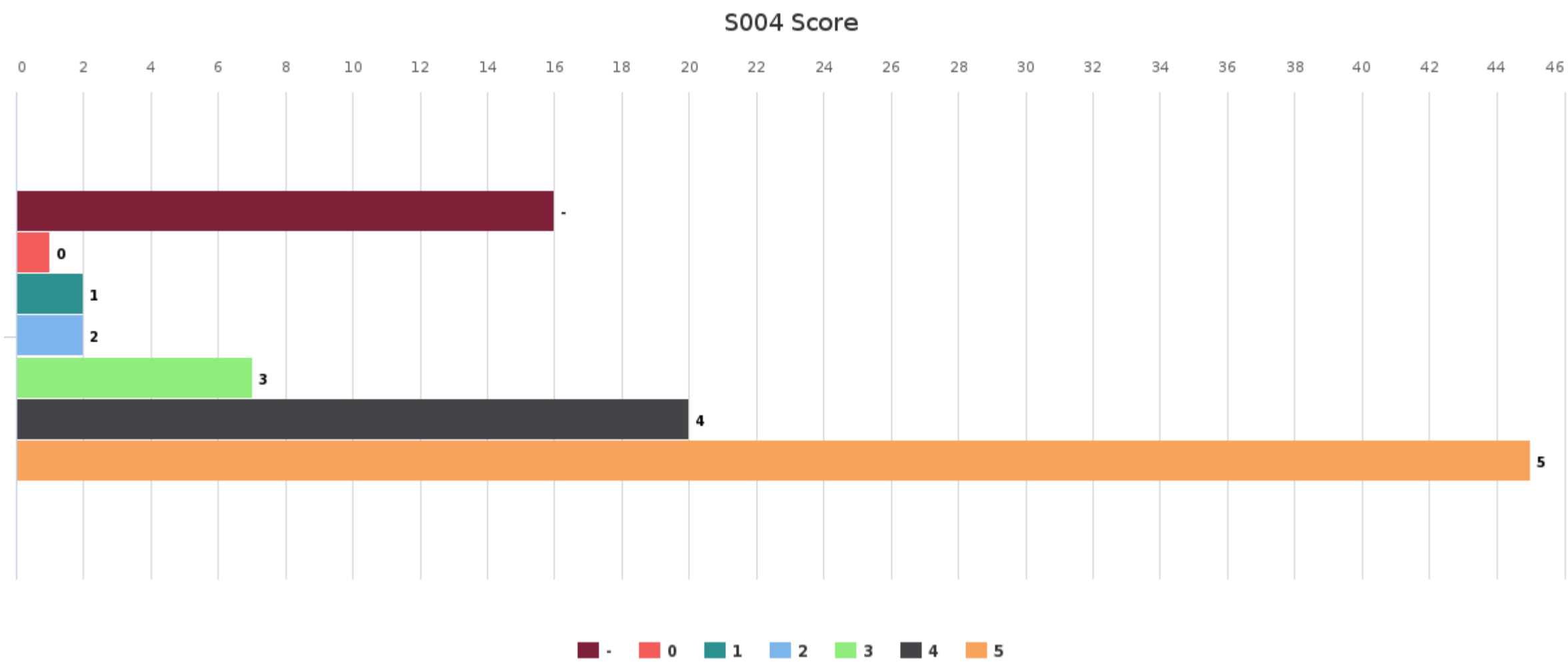


Chromogen: Instrument	Chromogen: Instrument count
Agilent Dako EnVision FLEX DAB	2
Agilent Dako EnVision FLEX DAB + Chromogen	14
Agilent Dako EnVision FLEX High pH HRP	7
Leica Biosystems Bond Polymer Refine Detection Kit	11
Other, please specify in comments field	1
Roche Ventana OptiView DAB IHC Detection Kit	11
Roche Ventana iVIEW DAB Detection Kit	2
Roche Ventana ultraView Universal DAB Detection Kit	20
Thermo Fisher Scientific UltraVision Quanto DAB	2
Total:	70



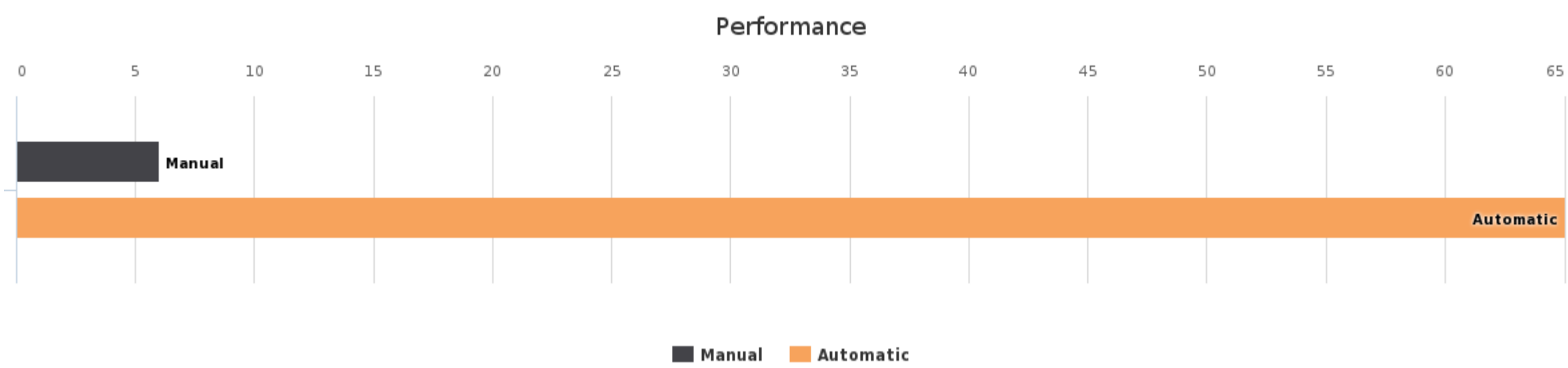
SCORE

Score	Comment
-	not available
0	false negative
1	almost negative staining, inadequate staining very weak staining, almost negative
2	understaining, unspesific staining weak staining
3	stain deposits, uneven, understained understained understained, uneven undertained, some background staining
4	dark hematoxylin, hematoxylin deposits slightly overstained slightly understained slightly understained, hematoxylin deposits slightly understained, stain deposits slightly understained, uneven slightly uneven some background staining stain deposits uneven, hematoxylin deposits uneven, some background staining uneven, stain deposits
5	Excellent hematoxylin deposits
Total:	



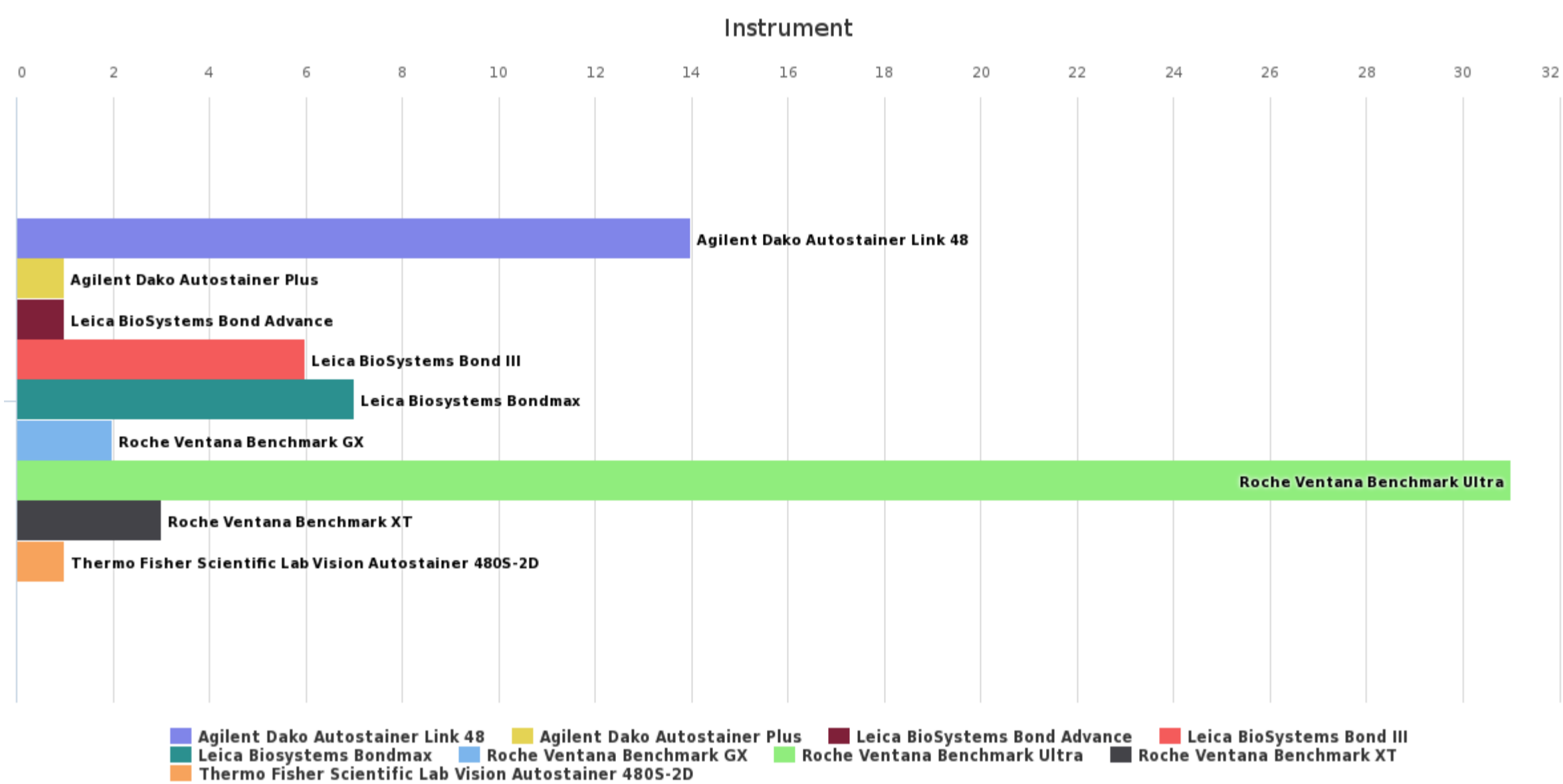
Score	Score count
-	16
0	1
1	2
2	2
3	7
4	20
5	45
Total:	93

S005 | Sample S005 PSA

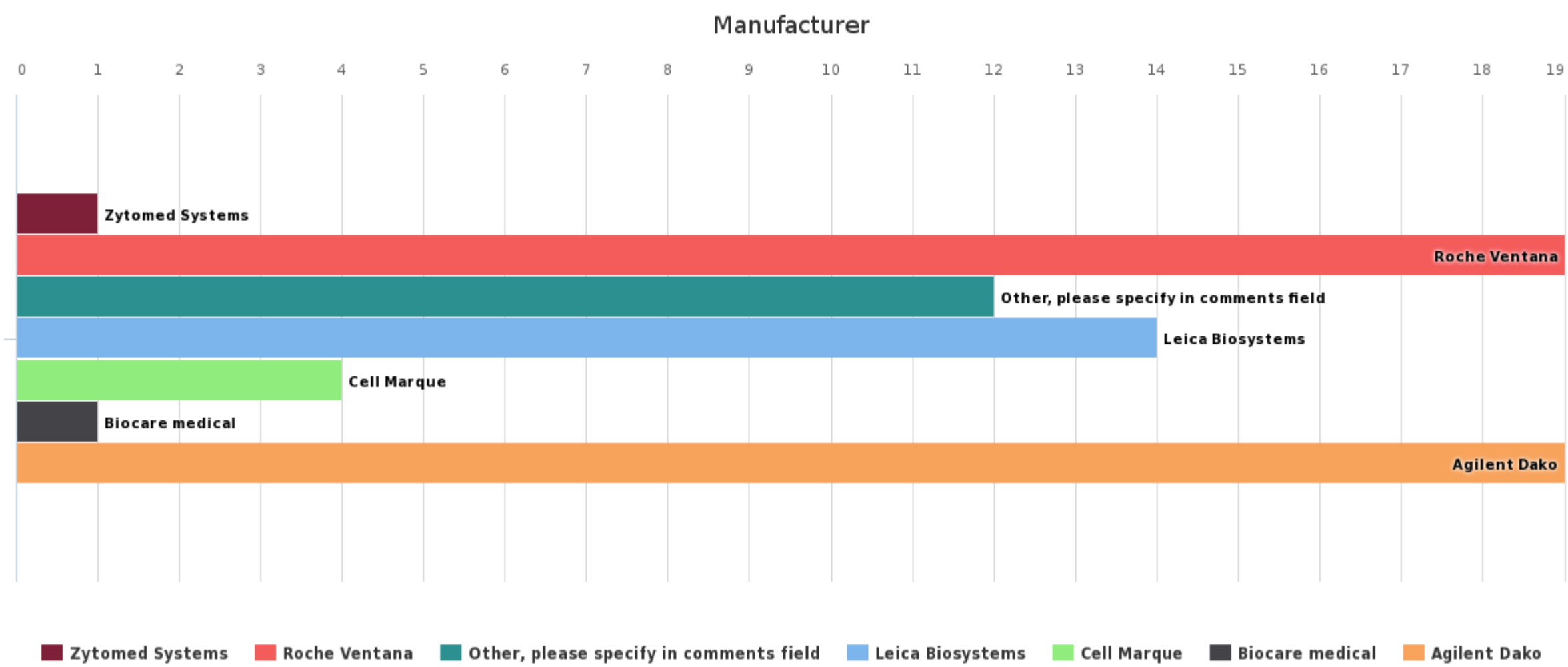


GENERAL DETAILS OF STAINING

Performance	Performance count
Automatic	65
Manual	6
Total:	71

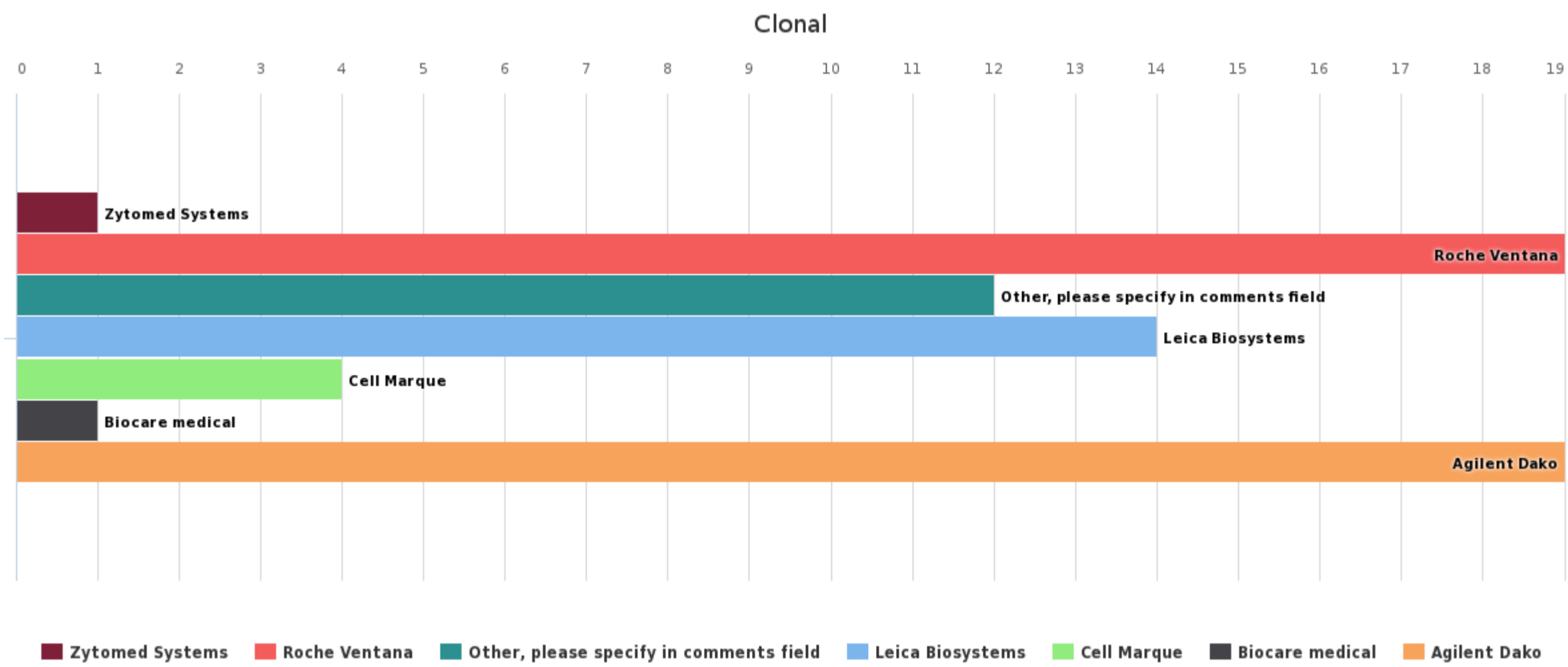


Instrument	Instrument count
Agilent Dako Autostainer Link 48	14
Agilent Dako Autostainer Plus	1
Leica BioSystems Bond Advance	1
Leica BioSystems Bond III	6
Leica Biosystems Bondmax	7
Roche Ventana Benchmark GX	2
Roche Ventana Benchmark Ultra	31
Roche Ventana Benchmark XT	3
Thermo Fisher Scientific Lab Vision Autostainer 480S-2D	1
Total:	66

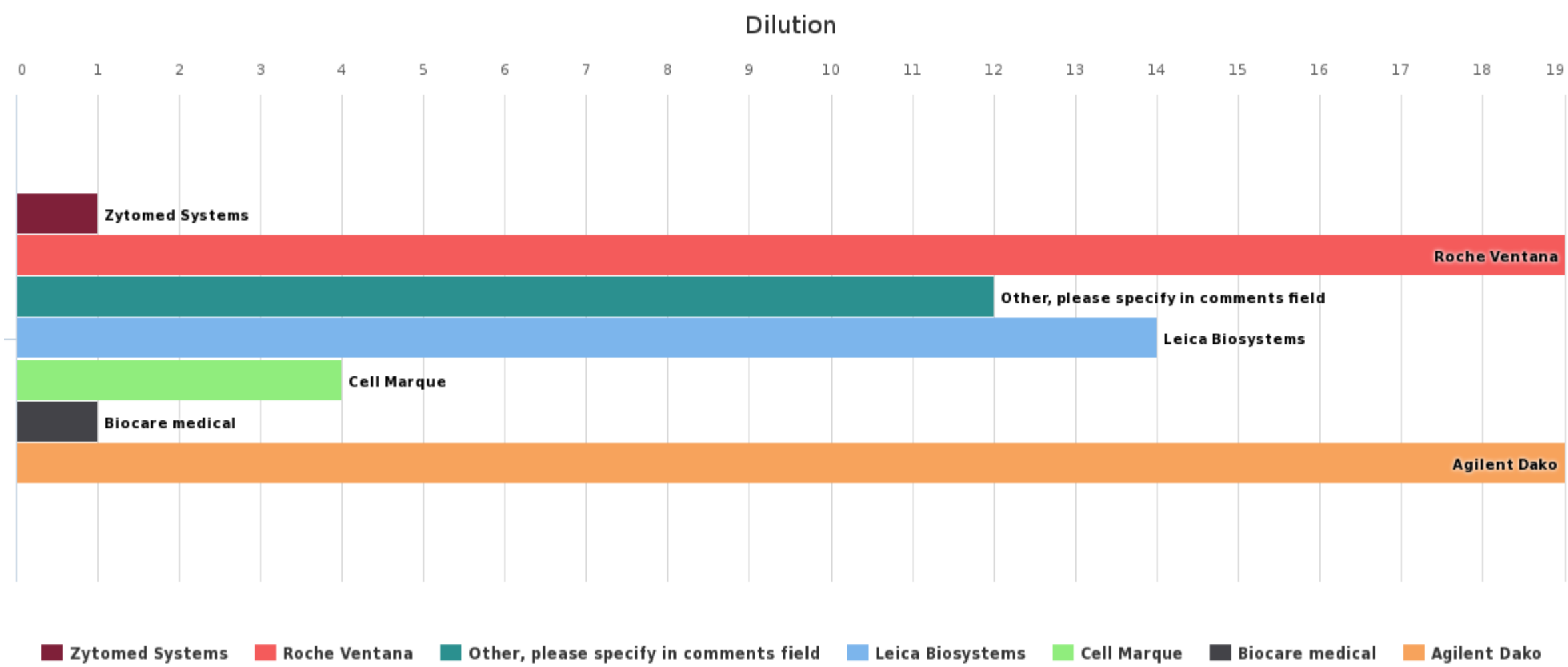


PRIMARY ANTIBODY

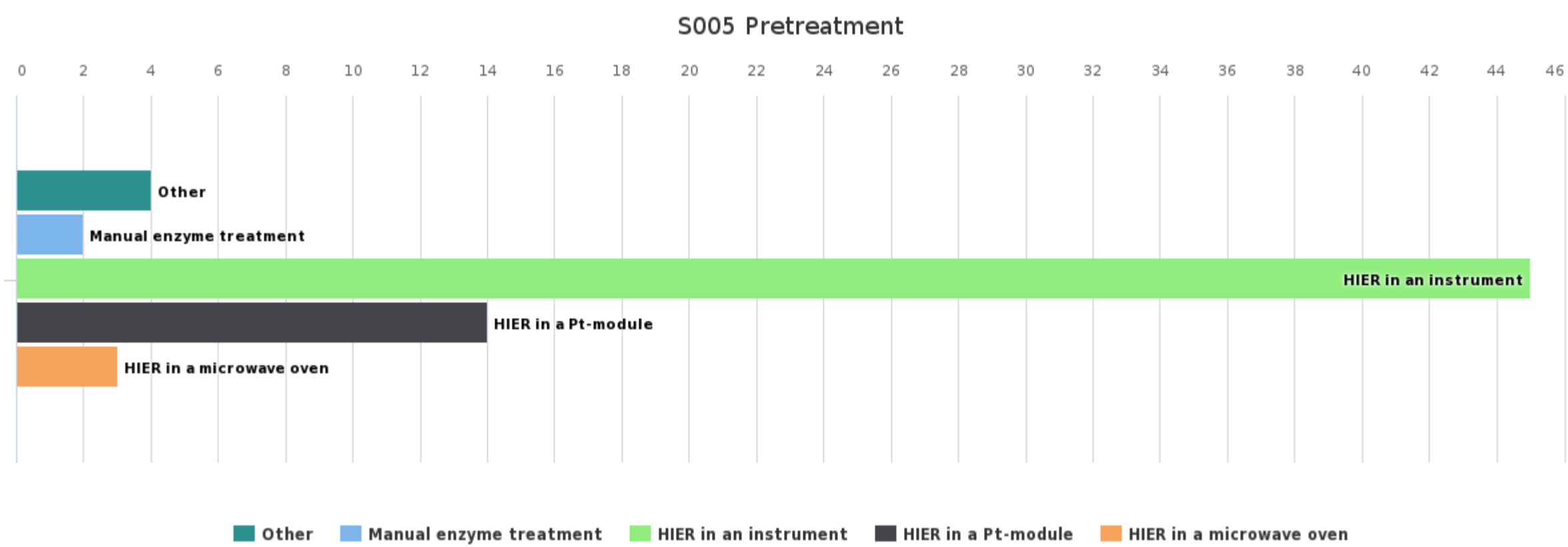
Manufacturer	Manufacturer count
Agilent Dako	19
Biocare medical	1
Cell Marque	4
Leica Biosystems	14
Other, please specify in comments field	12
Roche Ventana	19
Zytomed Systems	1
Total:	70



Clonal	Clonal count
Monoclonal	50
Polyclonal	19
Total:	69

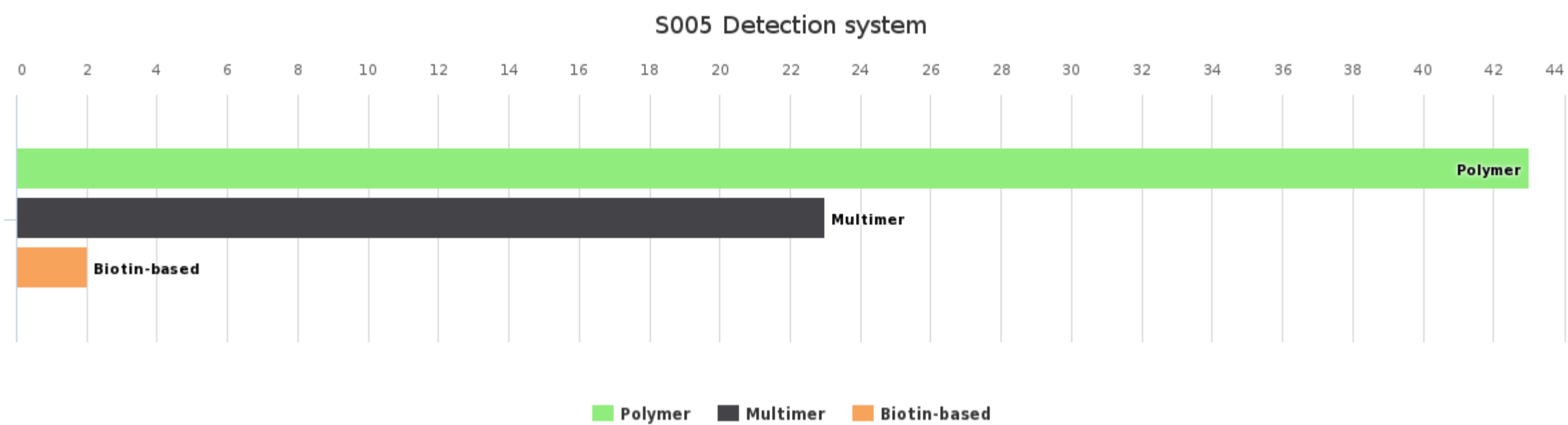


Dilution	Dilution count
Dilution	40
Ready-to-use	30
Total:	70



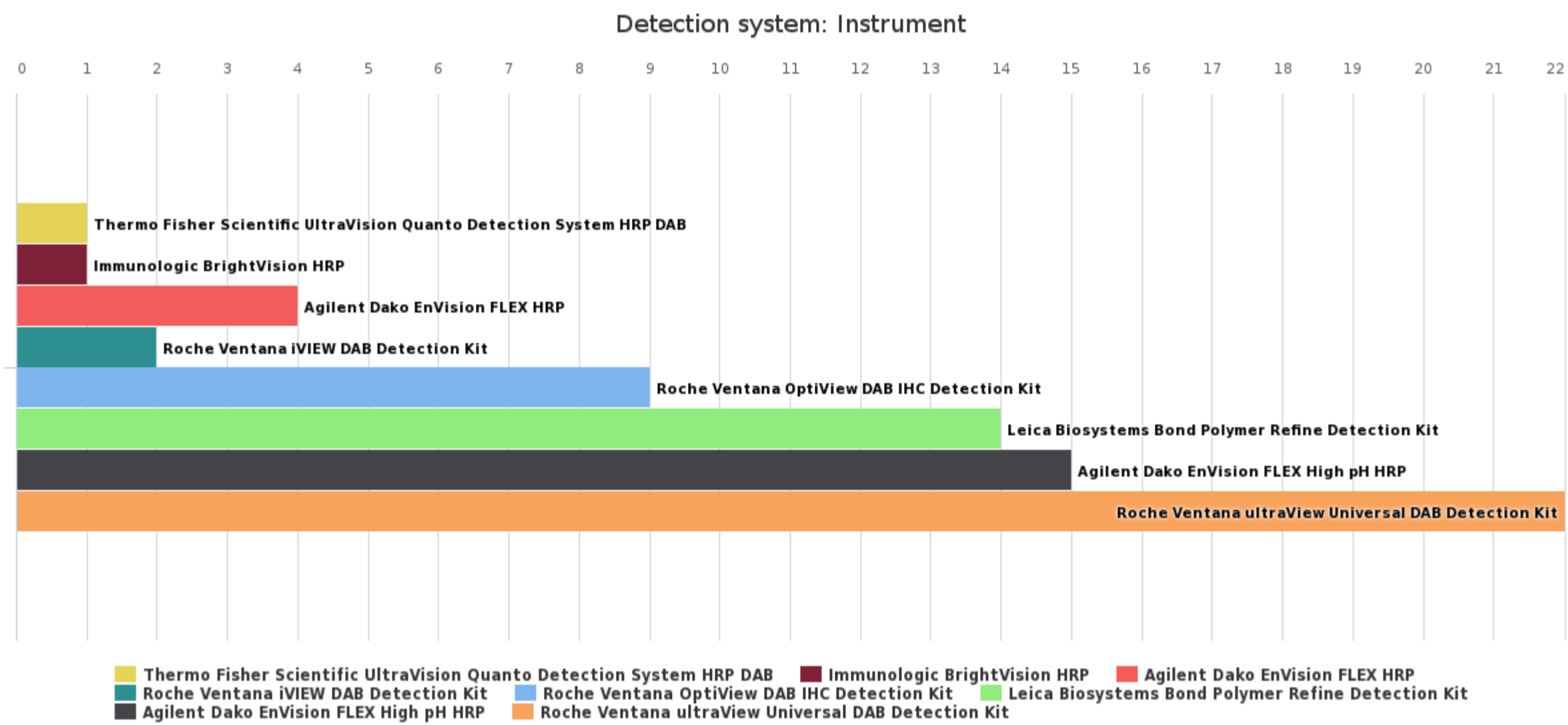
PRETREATMENT

Pretreatment	Pretreatment count
HIER in a microwave oven	3
HIER in a Pt-module	14
HIER in an instrument	45
Manual enzyme treatment	2
Other	4
Total:	68

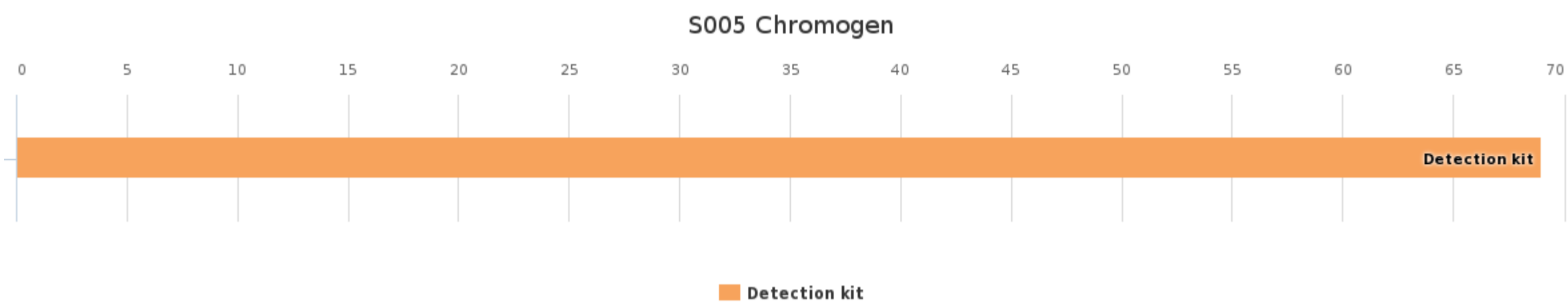


DETECTION SYSTEM

Detection system	Detection system count
Biotin-based	2
Multimer	23
Polymer	43
Total:	68

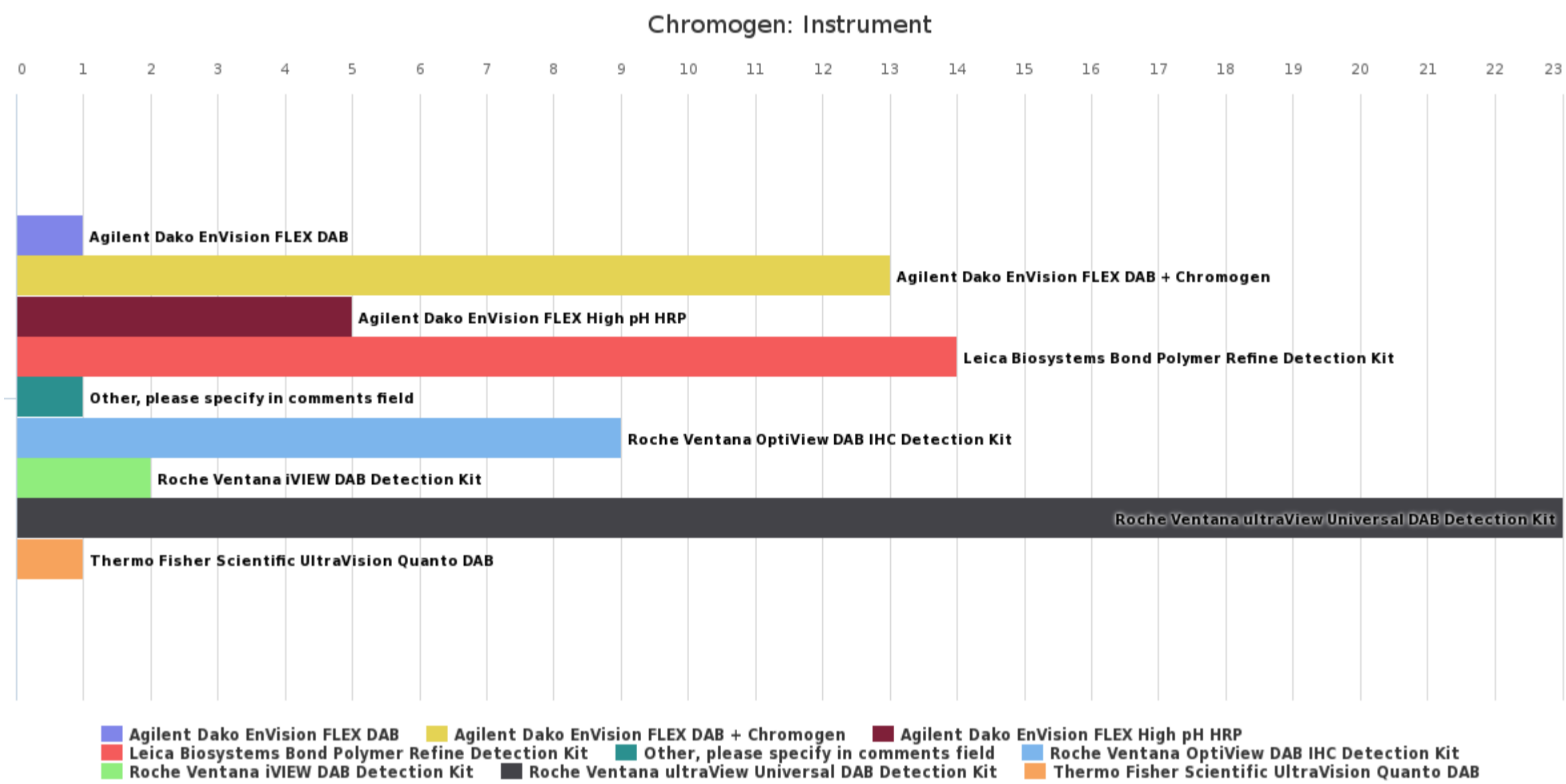


Detection system: Instrument	Detection system: Instrument count
Agilent Dako EnVision FLEX HRP	4
Agilent Dako EnVision FLEX High pH HRP	15
Immunologic BrightVision HRP	1
Leica Biosystems Bond Polymer Refine Detection Kit	14
Roche Ventana OptiView DAB IHC Detection Kit	9
Roche Ventana iVIEW DAB Detection Kit	2
Roche Ventana ultraView Universal DAB Detection Kit	22
Thermo Fisher Scientific UltraVision Quanto Detection System HRP DAB	1
Total:	68

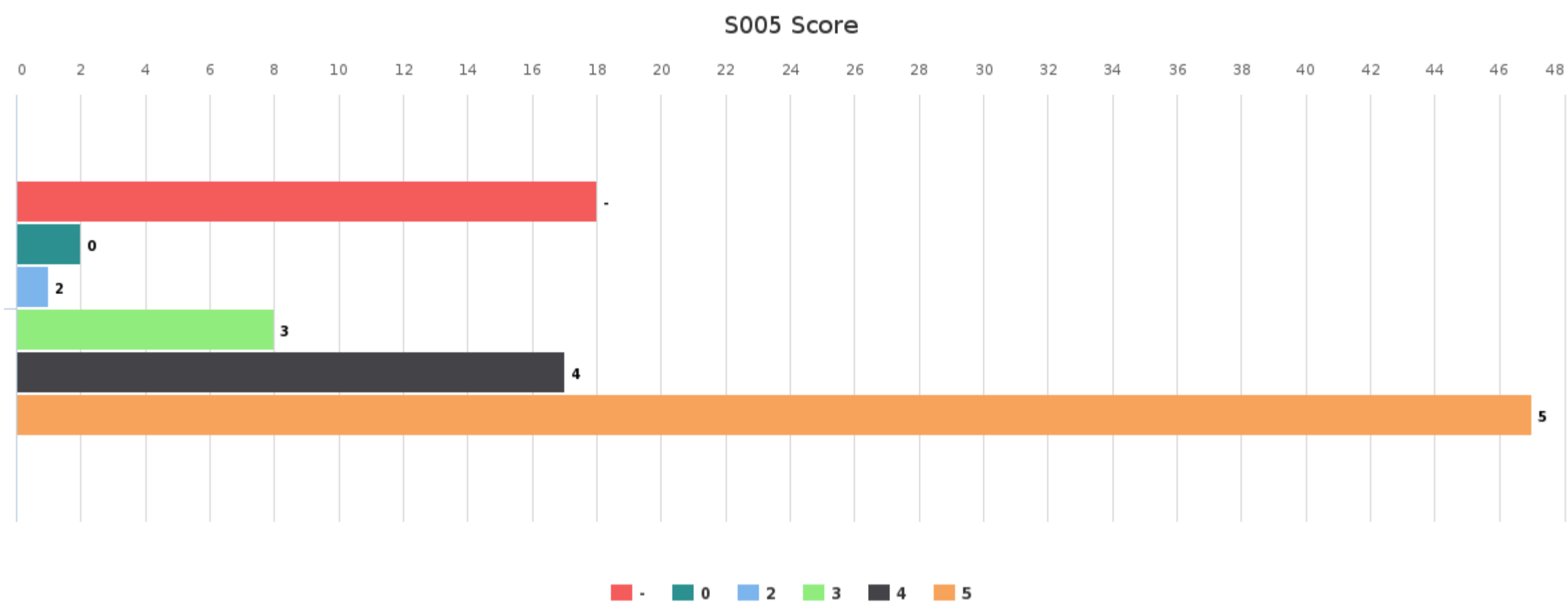


CHROMOGEN

Chromogen	Chromogen count
Detection kit	69
Total:	69

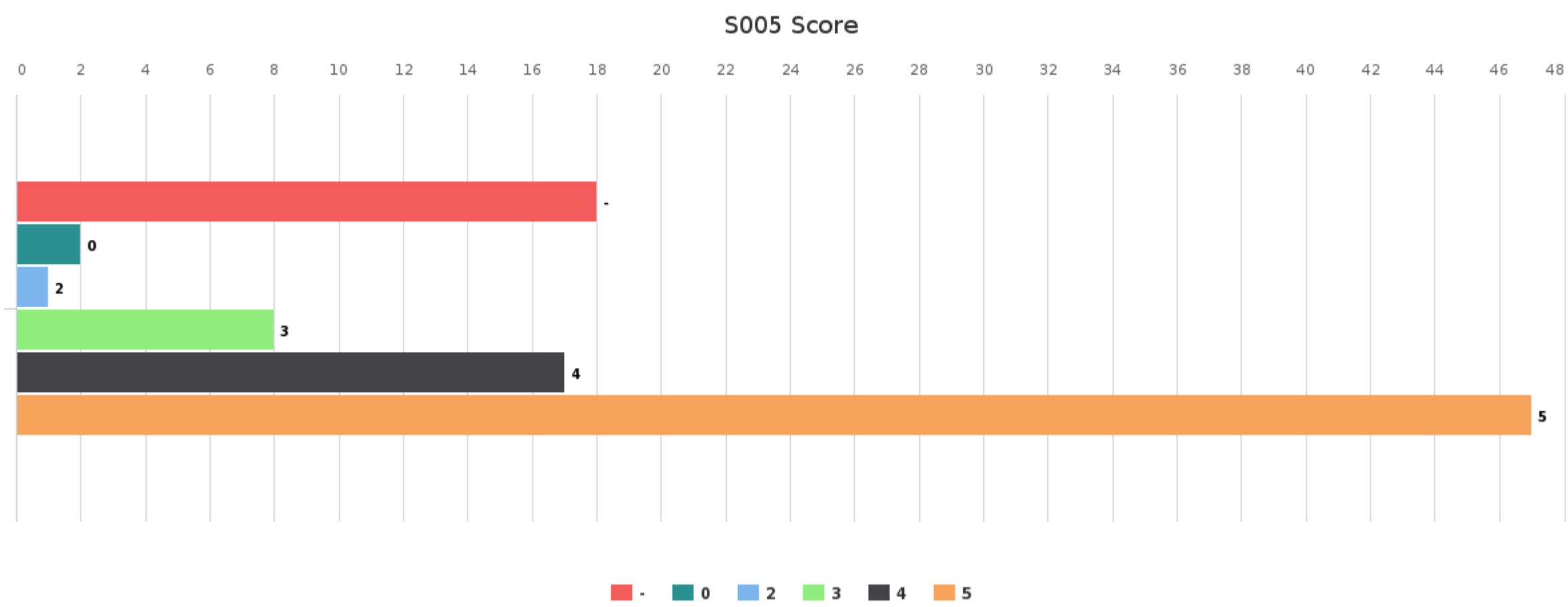


Chromogen: Instrument	Chromogen: Instrument count
Agilent Dako EnVision FLEX DAB	1
Agilent Dako EnVision FLEX DAB + Chromogen	13
Agilent Dako EnVision FLEX High pH HRP	5
Leica Biosystems Bond Polymer Refine Detection Kit	14
Other, please specify in comments field	1
Roche Ventana OptiView DAB IHC Detection Kit	9
Roche Ventana iVIEW DAB Detection Kit	2
Roche Ventana ultraView Universal DAB Detection Kit	23
Thermo Fisher Scientific UltraVision Quanto DAB	1
Total:	69



SCORE

Score	Comment
-	
	not available
0	
	negative
2	
	weak staining
3	
	dark hematoxylin, understained
	understained
	understained, dark hematoxylin
	understained, hematoxylin is quite weak
4	
	dark hematoxylin
	slightly understained
	slightly understained and uneven
	slightly understained, uneven
	slightly uneven
	uneven
5	
	Excellent
Total:	



Score	Score count
-	18
0	2
2	1
3	8
4	17
5	47
Total:	93

External Quality Assessment Scheme

## Immunohistochemical staining methods Round 3, 2022

### Specimens

Sample S001-S005 (LQ778322031-LQ778322035) were slides with unstained paraffin sections. The materials were sent without temperature control packaging.

### Report info

A numerical score given in a six-step scale 0–5 is based on consensus. The results of all participants are presented in a table form. A laboratory specific score report is enclosed as well. Scoring reports help your laboratory in following the development of your own stainings. Guidelines how to interpret the reports can be found under "LabScala user instructions" in LabScala. The scoring principles are presented below.

### Scoring

Evaluation scale: 0–5

3-5 points indicate good enough staining for diagnosis. 0-2 points mean that the staining is uncertain for diagnosis or failed.

5 points =	excellent = optimal
4 points =	practically faultless, slight over/understaining, slightly uneven or patchy staining
3 points =	good enough for diagnosis, but distinct over/understaining, uneven or patchy staining, stain deposits etc.
2 points =	weak staining, uncertain for diagnosis
1 point =	poor = failed, some scanty but inadequate staining observed/ notifiable overstaining leading to uncertain diagnosis, too much background, false positive or false negative
0 point =	negative or false positive - failed

### S001: CK7

#### Criteria for staining

At least weak cytoplasmic staining reaction on the majority on epithelial cells of the intercalating ducts in the pancreas. Epithelial cells of the large pancreatic ducts show a strong cytoplasmic staining reaction. A strong, distinct cytoplasmic staining reaction of bile duct epithelial cells in the liver. A strong cytoplasmic staining reaction in pancreatic adenocarcinoma.

### Results

CK 7 staining was evaluated from 88 laboratories. The average of the evaluated stainings was 4,4 points. The majority (97 %) of the stainings were evaluated as optimal (5 p.) or almost optimal (4 p.). One staining was evaluated as failed.

### Staining methods, instruments, and antibodies

About 93 % of laboratories used an IHC instrument for the staining and 7 % of laboratories stained samples manually. All laboratories used monoclonal antibodies. The most used clones were SP52 and OV-TL 12/30. Optimal staining results were reached also with the clone RN7. 61 % of laboratories used ready-to-use antibodies.

2023-02-21

### FINAL REPORT

Product no. 6600/6600S

Subcontracting: Sample preparation, sample pretesting

Samples sent	2022-11-01
Round closed	2022-12-13
Final report	2023-02-21

### Request for correction

Typing errors in laboratory's result forms are on laboratory's responsibility. Labquality accepts responsibility only for result processing. Requests must be notified by writing within three weeks from the date of this letter.

### Authorized by

EQA Coordinator  
Pia Eloranta  
pia.eloranta@labquality.fi

### Expert

Marjukka Friman,  
Medical cell biologist  
Heli Repo, Pathologist  
Wellbeing services county of Central Finland, Hospital Nova

### Labquality Oy

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FI-00520 HELSINKI  
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info@labquality.fi  
www.labquality.fi

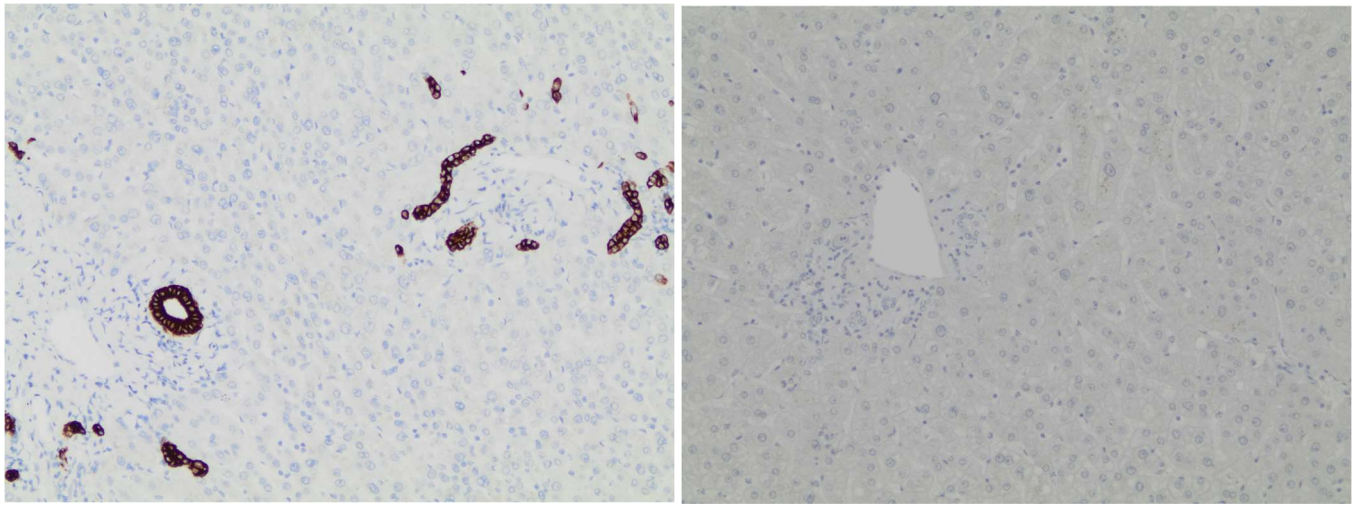


### **Pretreatments, incubation times and antibody dilutions**

Pretreatments were usually done with the IHC instrument or in the PT-module. Both alkaline and acid buffers were used as pretreatment. The details of the staining protocol (e.g pretreatment time, antibody dilution and antibody incubation time) varied widely.

### **Problems in CK7 -staining**

The most common defect was background staining.



Picture: Optimal and failed CK7 staining in kidney sample.

### **S002: WT1**

#### **Criteria for staining**

A strong nuclear staining of virtually all epithelial and smooth muscle cells in the fallopian tube. A moderate to strong nuclear staining of podocytes and parietal epithelial cells in kidney. A strong nuclear staining in metastatic sample.

#### **Results**

WT1 staining was evaluated from 72 laboratories. The average of the evaluated stainings was 3,9 points. The majority (76 %) of the stainings were evaluated as optimal (5 p.) or almost optimal (4 p). All stainings were evaluated as good enough for diagnosis (3-5 p.).

#### **Staining methods, instruments, and antibodies**

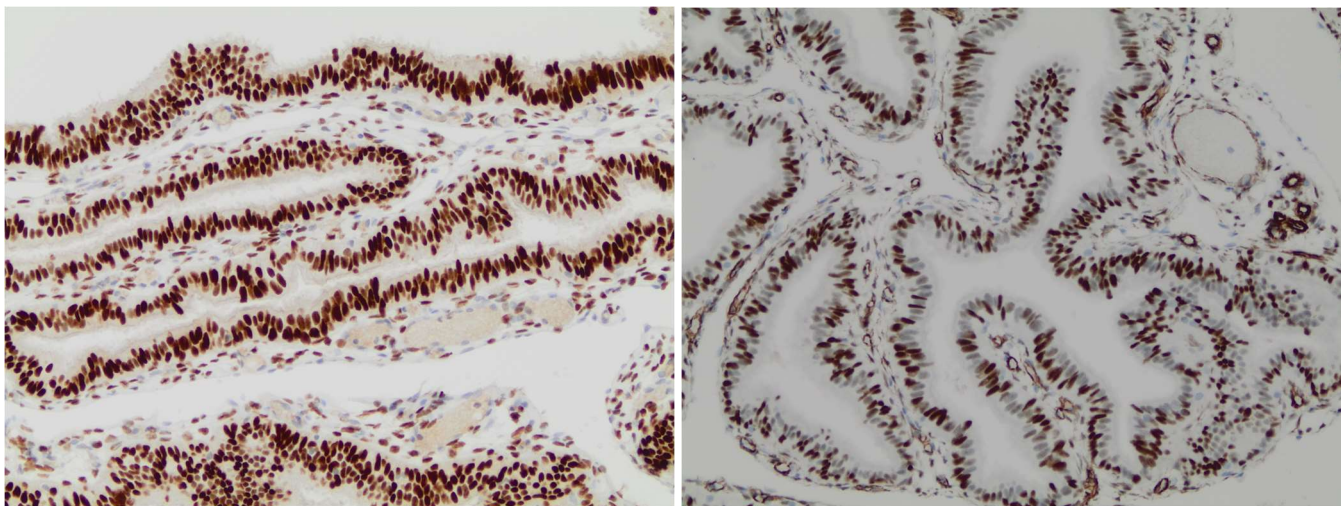
About 93 % of laboratories used an IHC instrument for the staining and 7 % of laboratories stained samples manually. All laboratories used monoclonal antibodies. The most used clones were WT49 and 6F-H2. Optimal staining results were reached with both clones. 67 % of laboratories used ready-to-use antibodies.

### **Pretreatments, incubation times and antibody dilutions**

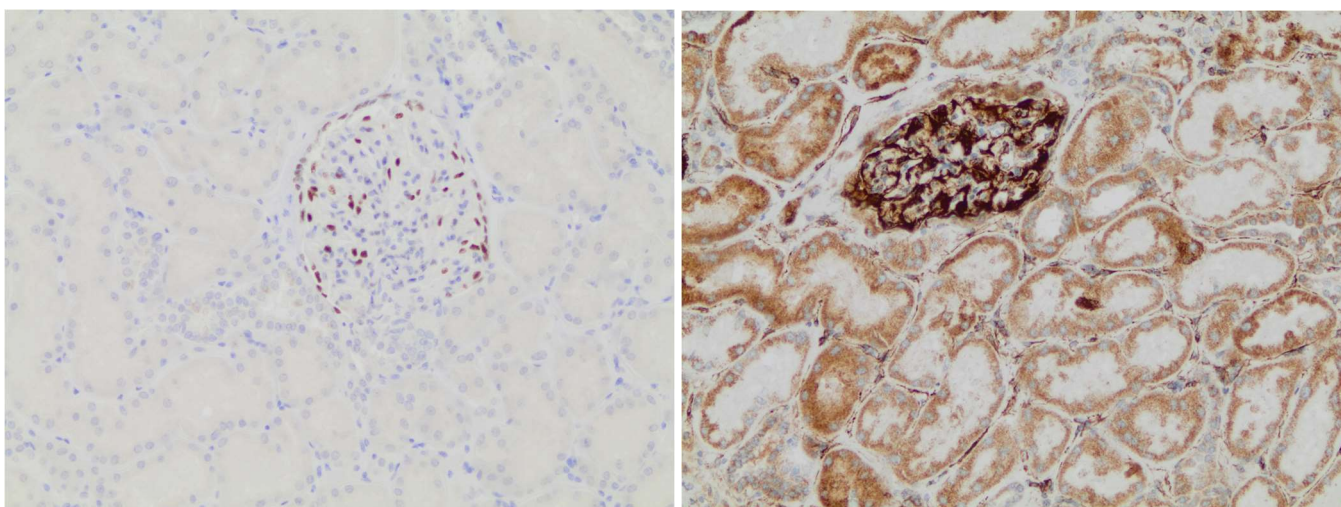
Pretreatments were usually done with the IHC instrument or in the PT-module. Alkaline buffers were used as pretreatment. The details of the staining protocol (e.g pretreatment time, antibody dilution and antibody incubation time) varied widely.

### **Problems in WT1 -staining**

The most common defects were understaining or overstaining.



Picture: Optimal and understained WT1 staining in fallopian tube sample.



Picture: Optimal and overstained WT1 staining in kidney sample.

### **S003: CK20**

#### **Criteria for staining**

A strong cytoplasmic staining reaction of epithelial cells in the colon and at least a weak to moderate staining reaction in most crypt cells. A strong cytoplasmic staining reaction in most of the colon adenocarcinoma cells.

#### **Results**

CK20 staining was evaluated from 84 laboratories. The average of the evaluated stainings was 4,3 points. The majority (82 %) of the stainings were evaluated as optimal (5 p.) or almost optimal (4 p). 4,8 % of stainings were evaluated as weak, poor or failed (0-2 p.).

#### **Staining methods, instruments, and antibodies**

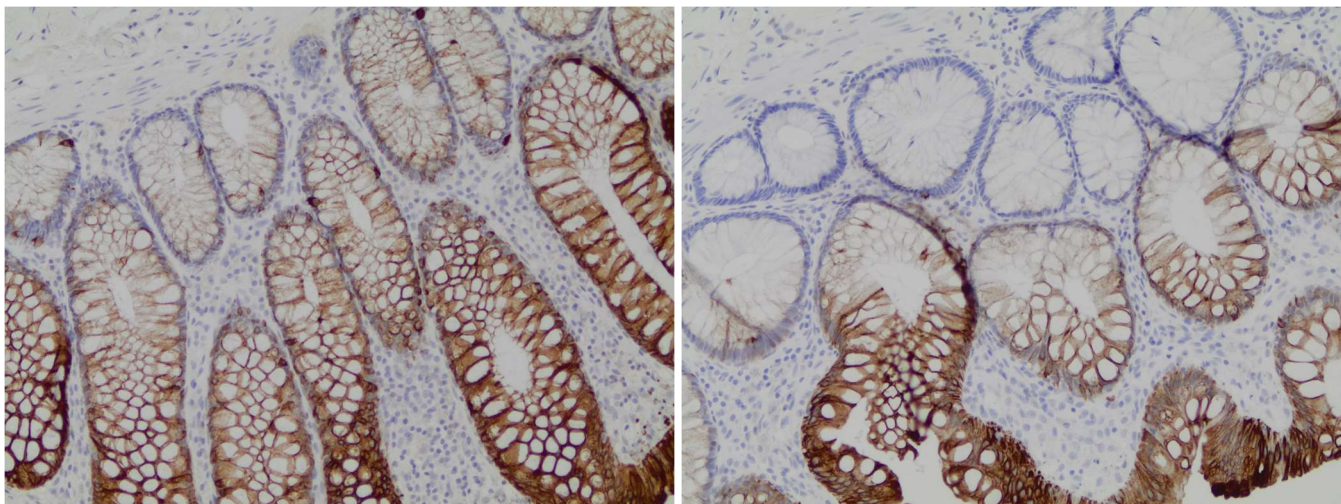
About 92 % of laboratories used an IHC instrument for the staining and 8 % of laboratories stained samples manually. About 99 % of laboratories used monoclonal antibodies. Used clones were SP33 and Ks20.8. Optimal staining results were reached with both clones. 58 % of laboratories used ready-to-use antibodies.

#### **Pretreatments, incubation times and antibody dilutions**

Pretreatments were usually done with the IHC instrument or in the PT-module. Both alkaline and acid buffers were used as pretreatment. The details of the staining protocol (e.g pretreatment time, antibody dilution and antibody incubation time) varied widely.

#### **Problems in CK20 -staining**

The most common defect was understaining.



Picture: Optimal and slightly understained CK20 staining in colon sample.

#### **S004: CDX2**

##### **Criteria for staining**

A strong nuclear staining reaction of epithelial cells in colon. A strong nuclear staining reaction of neoplastic cells in the colon adenocarcinoma.

##### **Results**

CK20 staining was evaluated from 77 laboratories. The average of the evaluated stainings was 4,3 points. The majority (82 %) of the stainings were evaluated as optimal (5 p.) or almost optimal (4 p). 6,5 % of stainings were evaluated as weak, poor or failed (0-2 p.).

##### **Staining methods, instruments, and antibodies**

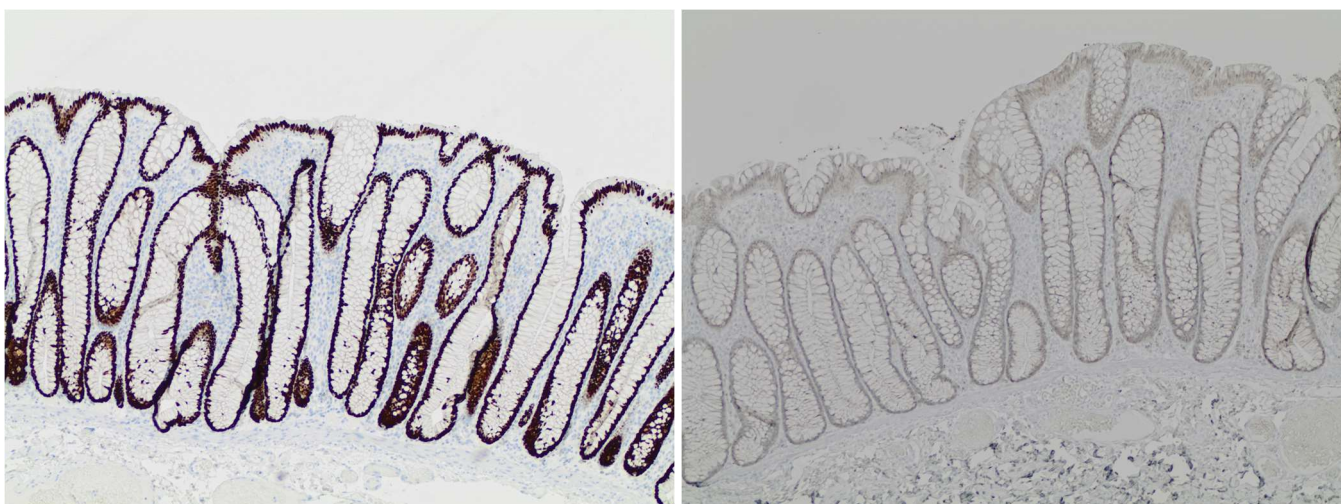
About 93 % of laboratories used an IHC instrument for the staining and 7 % of laboratories stained samples manually. About 99 % of laboratories used monoclonal antibodies. The most used clones were EPR2764Y and DAK-CDX2. Optimal staining results were reached also with clones EP25, IHC302 and CDX2-88. 62 % of laboratories used ready-to-use antibodies.

##### **Pretreatments, incubation times and antibody dilutions**

Pretreatments were usually done with the IHC instrument or in the PT-module. Alkaline buffers were used as pretreatment. The details of the staining protocol (e.g pretreatment time, antibody dilution and antibody incubation time) varied widely.

##### **Problems in CDX2 -staining**

The most common defects were understaining or background staining.



Picture: Optimal and weak CDX2 staining in colon sample.

## **S005: PSA**

### **Criteria for staining**

A strong cytoplasmic staining reaction of epithelial cells in the prostate glands. A strong cytoplasmic staining reaction of neoplastic cells in the prostate adenocarcinoma.

### **Results**

PSA staining was evaluated from 75 laboratories. The average of the evaluated stainings was 4,4 points. The majority (85 %) of the stainings were evaluated as optimal (5 p.) or almost optimal (4 p). 4,0 % of stainings were evaluated as weak, poor or failed (0-2 p.).

### **Staining methods, instruments, and antibodies**

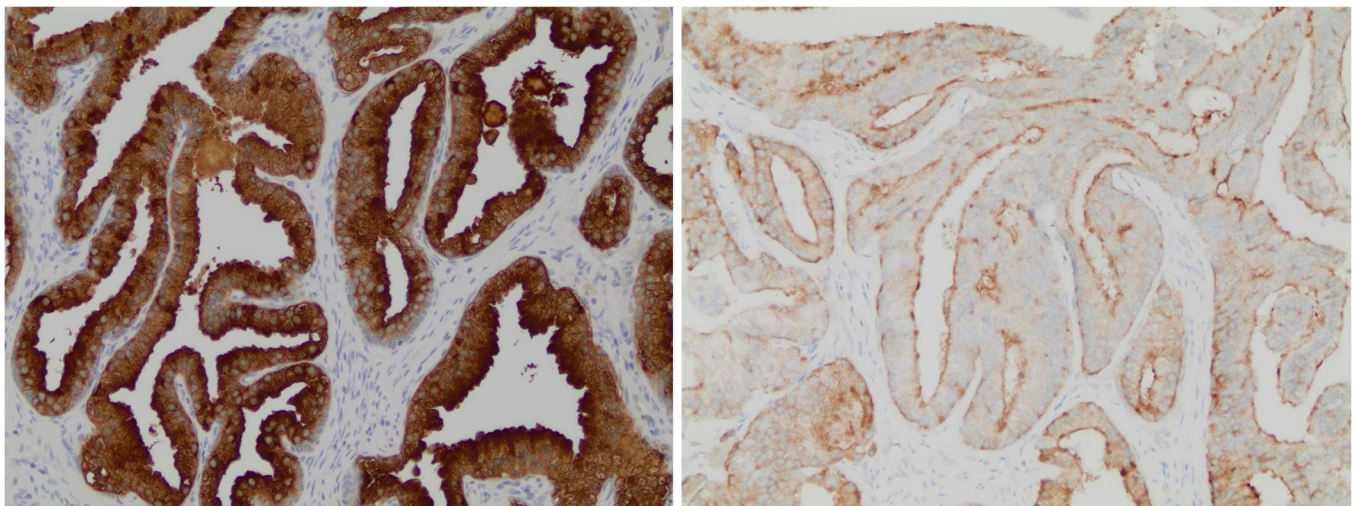
About 93 % of laboratories used an IHC instrument for the staining and 7 % of laboratories stained samples manually. About 72 % of laboratories used monoclonal antibodies and 28 % used polyclonal antibodies. The most used clones were ER-PR8 and 35H9. Optimal staining results were reached also with the clone IHC654 and with some polyclonal antibodies. 43 % of laboratories used ready-to-use antibodies.

### **Pretreatments, incubation times and antibody dilutions**

Pretreatments were usually done with the IHC instrument or in the PT-module. Both alkaline and acid buffers were used as pretreatment. The details of the staining protocol (e.g pretreatment time, antibody dilution and antibody incubation time) varied widely.

### **Problems in PSA staining**

The most common defect was understaining.



Picture: Optimal and weak PSA staining in prostate adenocarcinoma sample.

### **End of report**

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