

Technical Guide

Assays, Controls & Equipment



VAST 
Versatile Analyte Selection Technology

AccuBind 
ELISA Microwells

AccuLite 
CLIA Microwells



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Message from Monobind's president:



Dr. Frederick R. Jerome, CEO

Since its inception in 1977 Monobind, an ISO 13485 certified company, has pioneered in providing diagnostic tools to healthcare professionals at affordable prices. The expertise acquired through years of hard work has allowed Monobind significant immunoassay market penetration. With a distributor network spanning established and emerging worldwide markets in 110+ countries, Monobind looks forward to an even brighter future. As a global brand and leading manufacturer, our company is a major player in the field of diagnostic immunoassay systems.

Monobind's success can be attributed to the high quality and user friendliness of our product line. Customer care and quality consciousness are the hallmarks of Monobind. Close connections with universities and reference labs allow Monobind to provide the technical support to launch new projects in reduced time frames. Our experience in producing quality raw materials, for our own use and for other diagnostic manufacturers, delivers a sustainable competitive advantage built from cost-efficiency. Monobind's history

reveals a commitment to R&D and an impressive record of innovation in basic diagnostic techniques. Our products have helped transform the diagnostic practices from science to art by enabling improved, reliable patient care at an affordable cost.

In recent years, Monobind has integrated its systems to harmonize communications and work between R&D, QC, Manufacturing and Administration. We make continual improvements and have put substantial investment into our website to allow customers to access their account 24 x 7. Customers can go online to place orders, check order status, and view their account history and statements. Monobind's focus and dedication, as well as the market's growing demand for our product line make our many innovations possible.

This includes our totally unique, VAST® immunoassay product line which uses combination calibrators to support detection of multiple analytes in just one kit. VAST® is available in ELISA and CLIA for Anemia, Cancer, Diabetes, Fertility, Prenatal and Thyroid Panels with more to come in the future. Monobind also offers a range of QSure® Controls designed as a compliment to reagents and augmentation of laboratory QC.

Monobind works with leading automated and semi-automated instrument makers to customize equipment for our AccuBind® ELISA and AccuLite CLIA® products. Monobind offers analyzers for both technologies, including NeoEldex®, PrisMatic®, NeoLumax®, LuMatic™, and walkaway systems such as Autoplex® and TITIN® programmed to run our assays. Additionally, we provide 1000s of applications for existing instrumentation from such companies as Awareness Technology, Dynex and Gold Standard Diagnostics. We thank our partners for their collaboration and support, which has allowed our positioning to meet the needs of small-to-large volume testing in clinical labs.

Finally, I wish to thank our customers and distributors for their support and dedication. Their efforts have resulted in Monobind brand recognition in the highly competitive, global immunoassay market. This would not be possible without Monobind's team of skilled professionals who provide the fundamental base for our market success. With our rigorous pursuit of excellence and new product development, Monobind is building a robust future in the biomedical field. We hope to share this vision with you.

Dr. Frederick R. Jerome
CEO



Monobind Products

Our leading edge assay design, backed by substantial, ongoing R&D efforts, represents our fundamental commitment to deliver best-in-class products. To meet this goal several performance characteristics for our products were established and systems implemented throughout the organization to monitor it.

Most saliently, Monobind products must be easy-to-use, high quality, reproducible and stable. These apply to both ELISA and CLIA methodologies.

ELISA (Monobind AccuBind®) method is the mostly widely known and accepted design for diagnostic assays. Most assays have incubation times of 1 hour + 15 minutes with straight forward instructions. Competitive and equilibrium tests can be performed in this format on either antibody or streptavidin plates. Characteristic of this format is the colorimetric analysis using a spectrophotometer.

The second format of Monobind assays is CLIA (Monobind AccuLite®). New and state-of-the-art, CLIA assays offer some advantages compared to the traditional ELISA method. Competitive and equilibrium tests can still be performed in this format on either antibody or streptavidin plates but with shorter incubation times; the common incubation time is 45 minutes + 5 minutes. What truly distinguishes this format from any previous method is its use of a chemiluminescent compound to elicit the results of the reaction with an increased dynamic range and greater low-end sensitivity than ELISA.

Purpose of this Guide

Monobind wishes to bring user-friendly products and has created this guide to help ensure you have the best experience with our products. Knowing more about Monobind products and proper procedures, including common pitfalls is critical in developing your skill. This guide will deepen your understanding and help you perform the assays with excellent results time after time. It offers important information and tips directly from Monobinds' technical staff. You will learn how to interpret results and practice good laboratory technique.

We hope you enjoy this guide and your experience using Monobind products. Should you have any further questions, please contact your dealer or Monobind support staff at: techsupport@monobind.com

Product List

• Monobind AccuBind® ELISA and AccuLite® CLIA Kits

ALLERGY & ANEMIA

Ferritin Folate IgE – Immunoglobulin E	sTfR – Soluble Transferrin Receptor Vitamin B12
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BONE METABOLISM

Calcitonin PTH Intact	PTH Whole Vitamin D – 25-OH Total
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CANCER MARKERS

AFP – Alpha-Fetoprotein CA-125 Ovarian Cancer Antigen CA 15-3 Breast Cancer Antigen CA 19-9 Pancreatic Cancer Antigen CEA – Carcinoembryonic Antigen CEA NG – Carcinoembryonic Antigen Next Generation	fBhCG XR – Free Beta Human Chorionic Gonadotropin Extended Range fPSA – Free Prostate Specific Antigen tPSA – Total Prostate Specific Antigen tPSA XS– Total Prostate Specific Antigen Extra Sensitive
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CARDIAC MARKERS

CKMB – Circulating Creatine Kinase (MB) cTnI – Troponin I DIG – Digoxin	HS-CRP – High Sensitivity C-Reactive Protein Myoglobin
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DIABETES

C-Peptide	Insulin	Insulin Rapid
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ENDOCRINE

ACTH - Adrenocorticotropic Hormone
Aldosterone

INFECTIOUS DISEASE

Anti-H. Pylori IgA Anti-H. Pylori IgG Anti-H. Pylori IgM Anti-SARS-CoV-2 IgA	Anti-SARS-CoV-2 IgG Anti-SARS-CoV-2 S1-RBD IgG Anti-SARS-CoV-2 IgM D-Dimer	IL-6 Interleukin 6 PCT - Procalcitonin
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TORCH

Toxo IgG Toxo IgM Rubella IgG Rubella IgM	CMV IgG CMV IgM HSV 1+2 IgG HSV 1+2 IgM
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GROWTH DEFICIENCY

hGH – Human Growth Hormone

FERTILITY & PRENATAL

AMH FSH – Follicle Stimulating Hormone hCG – Human Chorionic Gonadotropin hCG R - Human Chorionic Gonadotropin Rapid hCG XR – Human Chorionic Gonadotropin Extended Range	LH – Luteinizing Hormone PAPP-A – Pregnancy Plasma Protein-A PRL – Prolactin PRLs – Prolactin Sequential
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NEONATAL

N-T4 – Neonatal Thyroxine N-17OHP – Neonatal 17-Alpha-Hydroxyprogesterone	N-TBG – Neonatal Thyroid Binding Globulin N-TSH – Neonatal Thyrotropin
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STERIODS

17OHP - 17-Alpha-Hydroxyprogesterone 17OHP-SI - 17-Alpha-Hydroxyprogesterone Scientific Unit ANST – Androstenedione Cortisol DHEA - Dehydroepiandrosterone DHEA-S – Dehydroepiandrosterone Sulfate E1 – Estrone	E2 – Estradiol Free Testosterone Progesterone SHBG – Sex Hormone Binding Globulin Testosterone uE3 – Estril
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THYROID

fT3 – Free Triiodothyronine fT4 – Free Thyroxine T3 – Triiodothyronine T3 R – Triiodothyronine Rapid T3 SBS– Triiodothyronine Streptavidin T3U – Triiodothyronine Uptake T4 – Thyroxine	T4 R – Thyroxine Rapid T4 SBS – Thyroxine Streptavidin TBG – Thyroxine-Binding Globulin Tg – Thyroglobulin TSH – Thyrotropin TSH R – Thyrotropin Rapid
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THYROID - AUTOIMMUNE

Anti-Tg – Anti-Thyroglobulin	Anti-TPO – Anti-Thyroperoxidase
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PrisMatic® ELISA Plate Analyzer
NeoEldex® ELISA Strip Analyzer
LuMatic® CLIA Plate Analyzer
NeoLumax® CLIA Strip Analyzer
Plate Wash Automated Microplate Washer



ASSAY SUMMARY - 1/3

DIAGNOSTIC AREA/ANALYTE	ITEM#	ASSAY TYPE	PLATE SYSTEM		BINDING		STEPS		SAMPLE VOL µl	SAM- PLE DILUTION	ELISA SUB	ELISA MANUAL INCUBATION TIME	CLIA MANUAL INCUBATION TIME
			Ab/Ag	STREPT	COMP	SAND	1-EQUIL	2-SEQU					

ALLERGY & ANEMIA

1	IgE	2500	4	-	S	-	Sa	-	2S	25	-	A + B	30+30+15 Min	30+30+5 Min
2	Ferritin	2800	4	-	S	-	Sa	-	2S	25	-	A + B	30+30+15 Min	30+30+5 Min
3	Folate	7500	8	-	S	C	-	1E	-	50	-	Sn	45+5 Min	45+5 Min
4	Folate & Vit B12	7800	8&9	-	S	C	-	1E	2S	50	-	Sn	Fol: 45+20 Min Vit B12: 45+30+20 Min	Fol: 45+5 Min Vit B12: 45+30+5 Min
5	sTFR	8600	4	-	S	-	Sa	-	2S	10	-	Sc	45+30+15 Min	30+30+5 Min
6	Vitamin B12	7600	9	-	S	C	-	2S	2S	50	-	Sn	45+30+20Min	45+30+5Min

BONE METABOLISM

7	Calcitonin	9300	2	Ab	-	-	Sa	1E	-	50	-	Sn	60+20 Min	45+5 Min
8	PTH Intact	9000	2	Ab	-	-	Sa	1E	-	50	-	Sn	60+20 Min	45+5 Min
9	PTH Whole & Intact	10000	2	Ab	-	-	Sa	1E	-	50 each	-	Sn	PTH Intact: 60+20 Min PTH Whole: 75+20 Min	PTH Intact: 45+5 Min PTH Whole: 60+5 Min
10	Vitamin D Direct	9400	6	Ab	-	C	-	-	2S	25	-	Sn	30+30+20 Min	30+30+5 Min

CANCER MARKERS

11	AFP	1900	3	-	S	-	Sa	1E	-	25	-	A + B	60+15 Min	45+5 Min
12	AFP, CEA, PSA	8400	3	-	S	-	Sa	1E	-	25 each	-	A + B	60+15 Min	45+5 Min
13	CA-125	3000	3	-	S	-	Sa	1E	-	25	-	A + B	60+15 Min	45+5 Min
14	CA 15-3	5600	4	-	S	-	Sa	-	2S	25	1:21	Sn	60+60+20 Min	30+45+5 Min
15	CA 19-9	3900	4	-	S	-	Sa	-	2S	25	-	A + B	60+60+15 Min	30+45+5 Min
16	CEA/CEA Nxt Gen	1800 4600	3	-	S	-	Sa	1E	-	25	-	A + B	60+15 Min	45+5 Min
17	fthCG	10200	4	-	S	-	Sa	-	2S	25	1:100	A + B	30+30+15 Min	30+30+5 Min
18	PSA/PSA XS	2100 8700	3	-	S	-	Sa	1E	-	25	-	A + B	30+15 Min	30+5 Min
19	Free PSA	2300	3	-	S	-	Sa	1E	-	50	-	A + B	60+15 Min	45+5 Min

CARDIAC MARKERS

20	DIG	900	7	-	S	C	-	1E	-	25	-	A + B	30+15 Min	30+5 Min
21	CKMB	2900	3	-	S	-	Sa	1E	-	25	-	A + B	15+15 Min	15+5 Min
22	cTnl	3800	2	Ab	-	-	Sa	1E	-	25	-	A + B	15+15 Min	15+5 Min
23	HS-CRP	3100	3	-	S	-	Sa	1E	-	25	1:200	A + B	15+15 Min	15+5 Min
24	Myoglobin	3200	3	-	S	-	Sa	1E	-	25	-	A + B	15+15 Min	15+5 Min

DIABETES

25	C-peptide	2700	3	-	S	-	Sa	1E	-	50	-	A + B	120+15 Min	60+5 Min
26	C-peptide & Insulin	7300	3	-	S	-	Sa	1E	-	50 each	-	A + B	120+15 Min	60+5 Min
27	Insulin	2400	3	-	S	-	Sa	1E	-	50	-	A + B	120+15 Min	60+5 Min
28	Insulin Rapid	5800	3	-	S	-	Sa	1E	-	50	-	Sn	60+15 Min	NA

ENDOCRINE

29	ACTH	10600	2	Ab	-	-	Sa	1E	-	50	-	Sn	60+20 Min	45+5 Min
30	Aldosterone	10100	9	-	S	C	-	-	2S	25	-	Sn	15+45+20 Min	15+45+5 Min





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DIAGNOSTIC AREA/ANALYTE	ITEM#	ASSAY TYPE	PLATE SYSTEM		BINDING		STEPS		SAMPLE VOL µl	SAM- PLE DILUTION	ELISA SUB	ELISA MANUAL INCUBATION TIME	CLIA MANUAL INCUBATION TIME
			Ab/Ag	STREPT	COMP	SAND	1-EQUIV	2-SEQU					

FERTILITY & PRENATAL

31	AMH	9700	2	Ab	-	-	Sa	1E	-	50	-	A + B	60+20 Min	60+5 Min
32	hCG	800	3	-	S	-	Sa	1E	-	25	-	A + B	60+15 Min	45+5 Min
33	hCG Ext. Range	8800	3	-	S	-	Sa	1E	-	25	-	A + B	20+15 Min	20+5 Min
34	hCG Rapid	3300	2	Ab	-	-	Sa	1E	-	25	-	SC	10+5 Min	NA
35	hCG, FSH, LH, PRLs	8300	3 & 4	-	S	-	Sa	1E	2S	hCG:PRLs: 25 LH/FSH: 50	-	A + B	hCG:20+15 Min LH/FSH:60+15 Min PRLs:30+30+15 Min	hCG:20+5 Min LH/FSH:45+5 Min PRLs:30+30+5 Min
36	FSH	400	3	-	S	-	Sa	1E	-	50	-	A + B	60+15 Min	45+5 Min
37	LH	600	3	-	S	-	Sa	1E	-	50	-	A + B	60+15 Min	45+5 Min
38	PAPP-A	12600	6	Ab	-	-	Sa	2S	2S	10	-	Sn	30 + 30 + 20 Min	30+30+5 Min
39	PRL	700	3	-	S	-	Sa	1E	-	25	-	A + B	60+15 Min	45+5 Min
40	PRL Sequ.	4400	4	-	S	-	Sa	2S	2S	25	-	A + B	30+30+15 Min	30+30+5 Min
41	AFP, hCG, uE3	8500	3 & 7	-	S	C	Sa	1E	-	25 each	-	A + B	60+15 Min	45+5 Min

GROWTH DEFICIENCY

42	hGH	1700	3	-	S	-	Sa	1E	-	50	-	A + B	60+15 Min	45+5 Min
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INFECTIOUS DISEASES

43	Anti-H. Pylori IgA	1600	1	-	S	-	Sa	-	2S	50	1:100	A + B	60+30+15 Min	45+30+5 Min
44	Anti-H. Pylori IgG	1400	1	-	S	-	Sa	-	2S	25	1:100	A + B	60+30+15 Min	45+30+5 Min
45	Anti-H. Pylori IgM	1500	1	-	S	-	Sa	-	2S	50	1:100	A + B	60+30+15 Min	45+30+5 Min
46	Anti-SARS-CoV-2 IgA	11800	10	Ag	-	-	Sa	-	2S	100	1:100	Sn	30+30+15 Min	30+30+5 Min
47	Anti-SARS-CoV2 IgG	11900	10	Ag	-	-	Sa	-	2S	100	1:100	Sn	30+30+15 Min	30+30+5 Min
48	Anti-SARS-CoV-2 S1-RBD IgG	12500	10	Ag	-	-	Sa	-	2S	100	1:100	Sn	30+30+15 Min	NA
49	Anti-SARS-CoV-2 IgM	11700	10	Ab	-	-	Sa	-	2S	100	1:100	Sn	30+30+20 Min	30+30+5 Min
50	D-Dimer	12000	4	Ab	-	-	Sa	-	2S	25	-	A + B	20+20+15 Min	20+20+5 Min
51	IL-6	12600	2	Ab	-	-	Sa	-	1E	50	-	Sn	80 Min	50 Min
52	PCT - Procalcitonin	9200	2	Ab	-	C	-	-	2S	50	-	Sn	30+15 Min	30+5 Min

TORCH

53	Toxo IgG	6100	10	Ag	-	-	Sa	-	2S	100	1:100	SC	75 Min	-
54	Toxo IgM	6200	10	Ag	-	-	Sa	-	2S	100	1:100	SC	75 Min	-
55	Rubella IgG	6300	10	Ag	-	-	Sa	-	2S	100	1:100	SC	75 Min	-
56	Rubella IgM	6400	10	Ag	-	-	Sa	-	2S	100	1:100	Sn	75 Min	-
57	CMV IgG	6500	10	Ag	-	-	Sa	-	2S	100	1:100	SC	75 Min	-
58	CMV IgM	6600	10	Ag	-	-	Sa	-	2S	100	1:100	Sn	75 Min	-
59	HSV 1+2 IgG	6700	10	Ag	-	-	Sa	-	2S	100	1:100	SC	75 Min	-
60	HSV 1+2 IgM	6800	10	Ag	-	-	Sa	-	2S	100	1:100	Sn	75 Min	-

NEONATAL THYROID & ADRENAL

61	N-TSH	3400	4	-	S	-	Sa	-	2S	DBS	-	Sn	90+45+45 Min (or OvrNt)	NA
62	N-T4	2600	6	Ab	-	C	-	-	2S	DBS	-	Sn	90+45+15 Min	NA
63	N-170HP	5500	7	-	S	C	-	1E	-	DBS	-	SC	30+90+15 Min Or 120+15 Min	NA
64	N-TBG	8900	9	-	S	C	-	-	2S	DBS	-	SC	60+30+30+15 Min (or OverNt)	NA





ASSAY SUMMARY - 1 / 3

DIAGNOSTIC AREA/ANALYTE	ITEM#	ASSAY TYPE	PLATE SYSTEM		BINDING		STEPS		SAMPLE VOL µl	SAM- PLE DILUTION	ELISA SUB	ELISA MANUAL INCUBATION TIME	CLIA MANUAL INCUBATION TIME
			AB/AG	STREPT	COMP	SAND	1-EQUIL	2-SEQU					

STERIODS

65	ANST	12400	5	Ab	-	C	-	1E	-	25	-	Sn	60+20 Min	45+5 Min
66	Cortisol	3600	7	-	S	C	-	1E	-	25	-	A + B	60+15 Min	45+5 Min
67	DHEA	7400	7	-	S	C	-	1E	-	25	-	Sn	60+20 Min	60+5 Min
68	DHEA-S	5100	7	-	S	C	-	1E	-	10	-	A + B	30+15 Min	30+5 Min
69	E1	10300	5	Ab	-	C	-	1E	-	25	-	Sn	45+20 Min	45+5 Min
70	E2	4900	9	-	S	C	-	-	2S	25	-	Sn	30+90+20 Min	30+60+5 Min
71	uE3	5000	7	-	S	C	-	1E	-	25	-	A + B	60+15 Min	45+5 Min
72	Progesterone	4800	7	-	S	C	-	1E	-	25	-	Sn	60+20 Min	60+5 Min
73	17OHP	5200	7	-	S	C	-	1E	-	25	-	Sn	60+15 Min	45+5 Min
74	170H-SI	9900	7	-	S	C	-	1E	-	25	-	SC	30+15 Min	45+5 Min
75	SHBG	9100	3	-	S	-	Sa	1E	-	25	-	A + B	30+15 Min	30+5 Min
76	Testosterone	3700	7	-	S	C	-	1E	-	10	-	A + B	60+15 Min	45+5 Min
77	Free Testosterone	5300	7	Ab	-	C	-	1E	-	20	-	A + B	60+15 Min	45+5 Min

THYROID

78	T3	100	5	Ab	-	C	-	1E	-	50	-	A + B	60+15 Min	45+5 Min
79	T3 Rapid	11200	5	Ab	-	C	-	1E	-	50	-	Sn	30+15 Min	NA
80	T3 SBS	8100	7	-	S	C	-	1E	-	50	-	A + B	60+15 Min	45+5 Min
81	T3, T4 & TSH	8000	7 & 3	-	S	C	Sa	1E	-	T3 / TSH : 50 T4 : 25	-	A + B	60+15 Min	45+5 Min
82	Free T3	1300	5	Ab	-	C	-	1E	-	50	-	A + B	60+15 Min	45+5 Min
83	FT3, FT4 & TSH	7000	7 & 3	-	S	C	Sa	1E	-	FT3: 50 FT4/TSH:25	-	Sn	60+15 Min	60+5Min
84	T3U	500	5	Ab	-	C	-	1E	-	25	-	A + B	60+15 Min	45+5 Min
85	T4	200	5	Ab	-	C	-	1E	-	25	-	A + B	60+15 Min	45+5 Min
86	T4 Rapid	11100	5	Ab	-	C	-	1E	-	20	-	A+B	30+15 Min	NA
87	T4 SBS	8200	7	-	S	C	-	1E	-	25	-	A + B	60+15 Min	45+5 Min
88	Free T4	1200	5	Ab	-	C	-	1E	-	50	-	A + B	60+15 Min	45+5 Min
89	TBG	3500	7	-	S	C	-	1E	-	10	-	A + B	30+15 Min	30+5 Min
90	Tg	2200	4	-	S	-	Sa	-	2S	50	-	Sn	1 Hr + 1 Hr + 15 Min or 1 Hr + 1 Hr + 5 Min or 2 Hr + 2 Hr + 15 Min 1.5 Hr + 1.5 Hr + 5 Min	
91	TSH	300	3	-	S	-	Sa	1E	-	50	-	A + B	60+15 Min	45+5 Min
92	TSH Rapid	6000	3	-	S	-	Sa	1E	-	25	-	A + B	30+15 Min	30+5

THYROID-AUTOIMMUNE

93	Anti-Tg	1000	1	-	S	-	Sa	-	2S	50	1:100	A + B	60+30+15 Min	30+30+5 Min
94	Anti-TPO	1100	1	-	S	-	Sa	-	2S	25	1:100	A + B	60+30+15 Min	30+30+5 Min

Abbreviation Key

PLATE SYSTEM		BINDING	STEPS		ITEM#
Ab	Antibody	C	Competitive	1E	1 Step Equilibrium
Ag	Antigen	Sa	Sandwich	2S	2 Step Equilibrium
S	Streptavidin	-	-	-	-

Contact us today to learn more

www.monobind.com

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TIPS FOR INTERPRETING ASSAY RESULTS

BAD DUPLICATION.

Possible Cause	Solution
Bubbles in wells.	Use a pin or needle to burst.
Dispensing error.	Check dispensing instrument.
Fingerprints/cell obstruction	Clean bottom surface of plate with DI water and dry before measuring again.
Improper wash step.	Ensure all wash equipment is working properly and wells are filled completely but do not overflow.

HIGH BACKGROUND.

Possible Cause	Solution
Insufficient wash step.	Repeat wash step. Ensure wells are filled completely but do not overflow. Consider increase in number of wash steps and soak time in between washes.
Contamination of reagents.	Run the test again with fresh reagents making sure to avoid contamination during dispensing.

LOW ABSORBANCE.

Possible Cause	Solution
Combined Substrate (A+B) not prepared correctly.	Prepare substrate reagent by mixing the correct volumes.
Contamination of reagents.	Run the test again with fresh reagents.
Temperature of the room may be lower than 20°C.	Increase the incubation time of the substrate but do not exceed 30 minutes.
Test volume is low.	Check pipette equipment for proper fit and calibration.
Increased plate moisture.	Ensure unused wells are sealed properly in pouch with desiccant and marked with open date.
Powerful wash step.	Reduce pressure of wash apparatus.
Analyzed test using incorrect wavelength.	Check that the equipment read wavelength is set to the same specified in the assay protocol.

CONTROLS VALUES ARE OUTSIDE OF ESTABLISHED VALUES.

Possible Cause	Solution
Contamination of controls.	Run the test again with new controls.
Contamination of calibrators.	Run the test again with new calibrators.
Incorrect control values.	Go to website for updated values.

PLATE STRIPS SLIP FROM HOLDER.

Possible Cause	Solution
Improper handling of plate.	When tapping, firmly grasp holder on grip marks.

PLATE STRIPS DO NOT FIT INTO HOLDER.

Possible Cause	Solution
Improper alignment of strips in holder. Incorrect holder for strip design.	Rotate strips 180° in holder. Use holder for strip design.

SPECIMENS GIVE ABSORBANCE OUTSIDE THE RANGE OF THE CALIBRATORS.

Possible Cause	Solution
Concentration of specimen is too high.	Dilute specimen with the “o” calibrator and run the assay again.



TIPS FOR INTERPRETING ASSAY RESULTS

SUBSTRATE “A” IS BLUE.

Possible Cause	Solution
The substrate is contaminated.	Obtain fresh Substrate “A”.

SUBSTRATE (A+B) TURNS BLUE WHEN MIXED.

Possible Cause	Solution
The substrate is contaminated.	Obtain fresh Substrate “A” and “B”.

AFTER THE STOP REAGENT IS ADDED, A BLUE-YELLOW COLOR REMAINS.

Possible Cause	Solution
This is a normal observation; the force of the stop reagent during addition did not cause sufficient mixing of the reagents.	Shake the plate, by hand or plate mixer, in order to sufficiently mix the reagents and a uniform yellow color results.

HIGH ABSORBANCE OF CALIBRATOR CAUSES OVERFLOW.

Possible Cause	Solution
The temperature of the room may be greater than 30°C or the plate was rotated during incubation.	Do not shake, rotate, or heat the plate during incubation. May measure the absorbance at 405nm because of the lower extinction constant.
Incubation time of substrate or reagent(s) step was longer than specified.	May measure the absorbance at 405nm because of the lower extinction constant.
The higher absorbance is not detrimental to the test but results may be limited by the analysis instrument.	

STOP SOLUTION IS YELLOW.

Possible Cause	Solution
Contamination of reagent.	Obtain fresh stop solution.

BEFORE MEASURING PLATE MORE THAN 30 MINUTES HAS LAPSED.

Possible Cause	Solution
Technician or instrument error.	Run the test again.
End product of enzyme reaction may precipitate and cause errors.	Run the test again.

HIGH ABSORBANCE OVER ENTIRE PLATE.

Possible Cause	Solution
Insufficient wash step.	Repeat wash step. Ensure wells are filled completely but do not overflow.
Contamination of substrate reagent.	Obtain fresh reagents and check pipette for proper dispensing.

POOR SENSITIVITY.

Possible Cause	Solution
Incorrect volume of reagent(s) added.	Check pipette for proper dispensing.
Time lapse between subsequent steps was longer than recommended.	Be sure to follow the test specifications as closely as possible to ensure accurate and reproducible results.



TIPS FOR INTERPRETING ASSAY RESULTS cont.

COLOR DEVELOPS QUICKLY.

Possible Cause	Solution
Contaminated reagents/equipment.	Ensure all equipment and reagents are free of contaminants then run the test again.

COLOR DEVELOPS SLOWLY.

Possible Cause	Solution
Reagents/samples are not at room temperature.	Do not run the assay until all reagents/samples are at room temperature.
Contamination of reagents/samples.	Only use the materials specified to avoid any cross reactions that may limit the reagents' activity within the assay.

POOR ASSAY REPRODUCIBILITY.

Possible Cause	Solution
Insufficient wash step.	Be sure that hand wash technique is consistent. If using an automatic plate washer, perform regular maintenance to ensure the machine is running properly and is clean.
Incubation temperature.	Try to maintain a consistent environment especially in regards to temperature. Refer to protocol for optimum reaction conditions.
Protocol Variations.	Be sure to follow the written protocol as closely as possible to limit the variation that may be introduced by the environment, equipment, and technician.

EDGE EFFECTS.

Possible Cause	Solution
Temperature of reaction vessel is uneven.	Maintain a consistent environment around the reaction vessel paying close attention to the temperature.

ASSAY DRIFT.

Possible Cause	Solution
Interruption(s) during assay set-up.	Set-up of assays should be free of interruptions and be done in the shortest time possible while maintaining good technique. To prevent any delays/interruptions while setting up an assay prepare all reagents and samples prior to beginning.
Reagents/samples are not at room temperature.	Dispense samples/reagents only when ALL are at room temperature to prevent variation of temperature throughout the plate wells.

AUTOMATION RESULTS ARE DIFFERENT THAN MANUAL METHOD.

Possible Cause	Solution
Automation application is not up to date.	Most current application for automation method should be downloaded from the website.



TIPS FOR GOOD LABORATORY TECHNIQUE

Microplates

The microplates are the scaffold on which the assay reaction proceeds so the correct type of plate is vital. When preparing for an assay determine whether an antibody or streptavidin plate will be required. Do not open and use any plate until it has reached room temperature. Once the pouch is opened, date the package, remove desired amount of strips, and seal remaining strips in the dated microplate pouch. Place the strips in the correct holder and place on a level surface for the remaining steps in assay setup. The strips should be inspected for any defects and should be cleaned of any residue using DI water.

Micro-pipetting

The volume used in assays is very small so it is extremely important to calibrate the pipettes accurately and in accordance with the time frame suggested by the manufacturer. In addition to calibration of the pipetting instrument, be sure to use the correct tips required for the specific pipette; the tips should consistently draw/ dispense the same volume and fit properly on the end of the pipette. To prevent contamination of any reagents used, change the pipette tip between dispensing of different samples/reagents. Further precaution can be taken by transferring only the amount of a sample/reagent needed to a separate container before dispensing. Two modes of dispensing can be performed, reverse and forward mode. Reverse mode is most recommended to use when dispensing controls/samples that need to be precise. Forward mode may be used when dispensing reagents that are in excess but this method is still not as accurate as the reverse mode.

Washing

The wash step is performed in order to remove the excess reagents from the well that now contains bound components. This step can either be done manually or with an automatic plate washer. The end result should be the same irrespective of the wash method used. The wash solution should flood each well 3-5 times and left to soak for 5-30 seconds between each wash, depending on the assay and the equipment being used. Many factors are taken into consideration when a wash solution's components are chosen so be sure to follow the suggested preparation. Here are some additional things to consider depending on the wash method of choice:

Manual – Fill each strip with the same volume of wash solution and keep the time it takes for each strip to be washed the same. No air bubbles should be trapped in the wells and the wells should not be overflowed with wash solution.

Plate Washers – Program the desired wash cycles and soak times for reproducible wash steps. Perform regular maintenance on the washers to ensure accurate dispensing volumes and that the wash heads are clean.

Substrate

In the line of Monobind products two different types of substrate exist. There are single component substrates and two component substrates. Depending on the assay different substrates may be used; Make sure to check the protocol and the components at hand for the correct reagents to use. Single component substrates are ready to use after attaining room temperature. Two component substrates need to be prepared/mixed prior to dispensing. Always check the substrates for contamination prior to use which is most often indicated by a blue coloration. Incubation time of the substrate is a vital point in the assay and time constraints should be kept according to those indicated in the protocol.



TIPS FOR GOOD LABORATORY TECHNIQUE

Conjugates

The conjugates used for each assay differ so be sure to use the correct conjugate for the assay. Storage of this component according to specifications is important because the storage and expiration of these components are established based on stability studies. When a dilution is required, only make the amount necessary for the immediate need. Before dispensing conjugates, they should be at room temperature and then stored at the specified conditions shortly after use.

Stop Solution

The last step of several assays involves the use of a stop solution. This component does exactly that, it stops the reaction from proceeding any farther. The solutions commonly used are acids at different molar concentrations. When running several assays always use the stop solution specified in the protocol for that assay. Once the stop solution has been added to the reaction mixture, there is a limited time period to analyze the plate and obtain the acceptable results. Reading results of the plate after 30 minutes has passed since addition of stop solution will result in data which may not be used with confidence.

Environment

To ensure reproducible results run all assays in a stable environment. An ideal stable temperature is one with little to no air drafts. In tropical or cold climates, it is recommended that labs regulate the room temperature (20 - 25 °C) to ensure proper reactions. Vibrations/rotations will also affect the test so these should be avoided. The surrounding environment is not the only of concern; the immediate environment contained in the plate holder is also critical.

All plates, samples/controls, conjugates, and other reagents should only be used when at room temperature. This will minimize any variations in temperature across the plate. A cover should always be placed on top to prevent any contamination or evaporation during incubation times. The main goal is to keep the environment as stable and consistent as possible.

Orders & Inquiries

Clinical Laboratories

Please place your orders through the Monobind Distributor in your region. If you do not know your regional distributor, you may contact Monobind for a referral.

Distributors

Existing distributors: please order online by logging into our web site using your account information, or send us a formal purchase order including the item catalog number, description, quantity required, the item rate, extended and total value.

New distributors: we welcome the opportunity to do business with you. To better understand and meet your diagnostic needs, please contact us with your product interest and distribution region. You may also visit us online and complete the New Customer Inquiry Form located in the Support Area of our web site.

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