WESTERN DETECTION

iBlot 3 Western Blot Transfer System

The next generation in performance and convenience

The Invitrogen™ iBlot™ 3 Western Blot Transfer System is an essential part of the Invitrogen™ western blotting workflow.

Electrophoresis

Protein gels welcome packs

Invitrogen™ PowerEase™ Touch power supplies

Transfer

iBlot 3 Western Blot Transfer System

Processing

Invitrogen™ iBind™ and iBind™ Flex automated western processing devices

Primary and secondary antibodies

Imaging

Invitrogen™ iBright™ Imaging Systems

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The iBlot 3 Western Blot Transfer System is a next-generation dry protein transfer solution with ready-to-use consumables that delivers high performance and convenience. The Invitrogen™ iBlot™ 3 Western Blot Transfer Device helps maximize laboratory productivity, enables excellent transfer efficiency, and helps provide consistent and reproducible results.

Maximum productivity

Perform reliable, high-quality transfers in as few as 3 minutes. Save on the time usually spent waiting for a transfer device to become available—the two independently controlled transfer stations of the iBlot 3 system allow you to share the device, simultaneously running a total of four mini gels or two midi gels with two different programs. Reduce protocol setup time by selecting preprogrammed protocols from the user-friendly interface.

Figure 1. The all-new design of the iBlot 3 device features two independently controlled transfer stations with programmable cooling.

Figure 2. Four mini transfer stacks loaded in the iBlot 3 device.

Better transfer efficiency

With the iBlot 3 device, you can achieve equivalent or better protein transfer efficiency compared with wet-tank transfer and other rapid transfer methods. The new design offers unique built-in adjustable cooling so that you can optimize the transfer of both high- and low-molecular weight proteins.

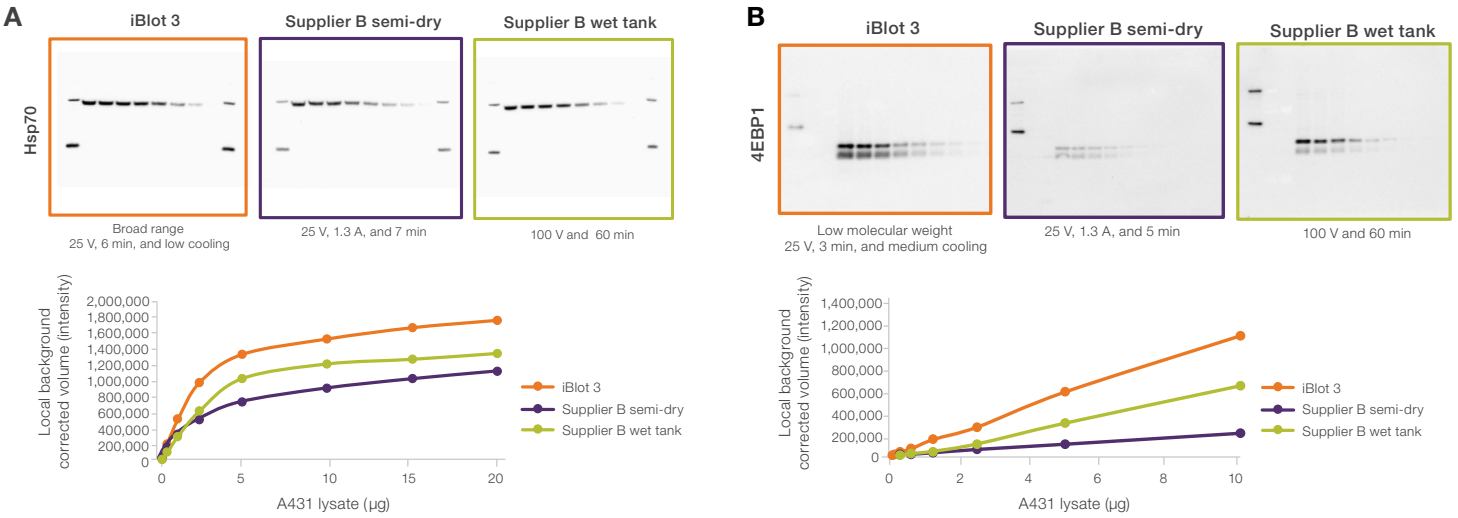

Figure 3. The iBlot 3 system demonstrates superior transfer efficiency. (A) Hsp70: Invitrogen™ NuPAGE™ 4 to 12% Bis-Tris mini gels were loaded with 20, 15, 10, 5, 2.5, 1.25, 0.625, and 0.312 µg of A431 lysate per lane, respectively. After gel electrophoresis, the proteins were transferred using the method and conditions shown above. Immunoprocessing was completed using the Invitrogen™ Bandmate™ Automated Western Blot Processor. Blots were incubated with Thermo Scientific™ Pierce™ Clear Milk Blocking Buffer for 30 minutes. Hsp70 primary antibody (1:1,000 in clear milk) was added to blots and incubated for 12 hours at room temperature. Primary antibody was removed and GAM-HRP secondary antibody (1:120,000 in TBST) was added to blots and incubated for 2 hours. Thermo Scientific™ SuperSignal™ West Dura chemiluminescent substrate was used for detection. The Invitrogen™ iBright™ FL1500 Imaging System was used for image capture. Local background corrected volume per lysate was plotted for each lane. **(B)** 4EBP1: Invitrogen™ Novex™ 16% Tricine mini gels were loaded with 10, 5, 2.5, 1.25, 0.625, 0.312, 0.156, and 0.078 µg of A431 lysate per lane, respectively. After gel electrophoresis, the proteins were transferred using the method and conditions shown above. Immunoprocessing was completed using the Bandmate Automated Western Blot Processor. Blots were incubated with Pierce Clear Milk Blocking Buffer for 30 minutes. 4EBP1 primary antibody (1:1,000 in clear milk) was added to blots and incubated for 12 hours at room temperature. Primary antibody was removed and GAR-HRP secondary antibody (1:175,000 in TBST) was added to blots and incubated for 2 hours. SuperSignal West Dura chemiluminescent substrate was used for detection. The iBright FL1500 instrument was used for image capture. Local background corrected volume per lysate was plotted for each lane. Note: GAM = goat anti-mouse, HRP = horseradish peroxidase, TBST = tris-buffered saline with Tween™ (solution), GAR = goat anti-rabbit.

Consistent, reproducible results

The preassembled single-use consumables and ultrafast transfer methods reduce variability—helping ensure consistent and repeatable results compared with classical methods. In addition, the all-new robust design with built-in cooling reduces heat buildup and enables improved run-to-run consistency.

Figure 4. Invitrogen™ iBlot™ 3 Transfer Stacks. Each box contains 10 stacks (mini or midi format), 10 filter papers, and 10 absorbent pads.

How it works

The short distance between electrodes, along with high field strength and current, reduces transfer time to as few as 3 minutes.

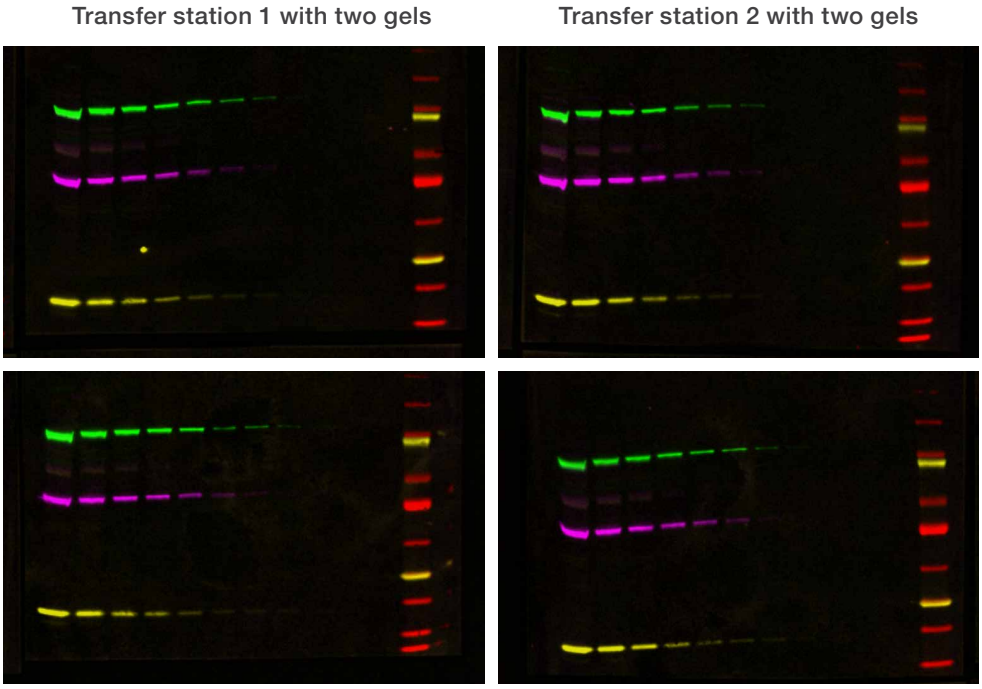

Figure 5. The iBlot 3 device delivers consistent results. A serial dilution of HEK293 lysate (lysed in Thermo Scientific™ RIPA buffer) was loaded onto four Invitrogen™ Bolt™ 4 to 12% Bis-Tris Plus gels. Protein loads were (left to right, in µg): 10, 5, 2.5, 1.25, 0.625, 0.312, 0.156, 0.078, 0.039, 0.020, and 0.010. Invitrogen™ iBright™ Prestained Protein Ladder was loaded into the last lane. MES-SDS running buffer was used for gel electrophoresis. Proteins were transferred using the iBlot 3 Western Blot Transfer Device and iBlot 3 Transfer Stacks, Midi, NC using the broad range (30–250 kDa) preprogrammed transfer method (25 V, 6 min, low cooling). Blots were then blocked with Pierce Clear Milk Blocking Buffer and were then probed using primary antibodies raised against epidermal growth factor receptor (EGFR), calreticulin, and p23 in clear milk for 12 hours. Secondary antibodies Invitrogen™ GAR-Alexa Fluor™ Plus 800 dye, Invitrogen™ GAM-Alexa Fluor™ Plus 488 dye, and Invitrogen™ GAChk-Alexa Fluor™ 546 dye were prepared in TBST and added to the blots for 2 hours. Immunoprocessing was completed on the Bandmate Automated Western Blot Processor. Imaging was completed on the iBright FL1500 Imaging System. Note: GAChk = goat anti-chicken.

Ordering information

Description	Quantity	Cat. No.
iBlot 3 Western Blot Transfer Device	1	IB31001
iBlot 3 Western Blot Transfer Device with Extended Warranty	1	A56727
iBlot 3 Transfer Stacks, Midi, NC	10 stacks	IB33001
iBlot 3 Transfer Stacks, Mini, NC	10 stacks	IB33002
iBlot 3 Transfer Stacks, Midi, PVDF	10 stacks	IB34001
iBlot 3 Transfer Stacks, Mini, PVDF	10 stacks	IB34002
1-Year Rapid Exchange Extended Warranty	1 year	ZGEXSCIBLOT3
1-Year ABRC Support Extended Warranty	1 year	ZG03SCIBLOT3
1-Year ABRC Support Plus Extended Warranty	1 year	ZG04SCIBLOT3
iBlot 3 Starter Kit, NC	1 kit	IB31001S
iBlot 3 Starter Kit, PVDF	1 kit	IB31002S
iBlot 3 Starter Kit, NC with Extended Warranty	1 kit	A56728
iBlot 3 Starter Kit, PVDF with Extended Warranty	1 kit	A56729
iBlot 3 iBind/iBind Flex Starter Kit*	1 kit	IB31001SLF1/ IB31001SLF2
Bolt Welcome Pack with iBlot 3 system	1 pack	NW0412AIB3
Starter kit and welcome pack contents		
iBlot 3 Starter Kit, NC		
iBlot 3 Western Blot Transfer Device	1	IB31001
iBlot 3 Transfer Stacks, Midi, NC	2 boxes (20 transfer stacks)	IB33001
iBlot 3 Starter Kit, PVDF		
iBlot 3 Western Blot Transfer Device	1	IB31001
iBlot 3 Transfer Stacks, Midi, PVDF	2 boxes (20 transfer stacks)	IB34001
iBlot 3 iBind Starter Kit		
iBlot 3 Western Blot Transfer Device	1	IB31001
iBlot 3 Transfer Stacks, Midi, NC	1 box (10 transfer stacks)	IB33001
iBind Western Device	1	SLF1000
iBind Cards	1	SLF1010
iBind Solution Kit	1	SLF1020
SuperSignal West Pico Plus Chemiluminescent Substrate	200 mL	34577
Bolt Welcome Pack with iBlot 3 system		
iBlot 3 Western Blot Transfer Device	1	IB31001
iBlot 3 Transfer Stacks, Midi, NC	1 box (10 transfer stacks)	IB33001
Mini Gel Tank	1	A25977
20X Bolt MES SDS Running Buffer	500 mL	B0002
4X Bolt LDS Sample Buffer	10 mL	B0007
10X Bolt Sample Reducing Agent	10 mL	B0009
PageRuler Plus Prestained Protein Ladder, 10–250 kDa	2 x 250 µL	26619
Bolt 4 to 12% Bis-Tris 1.0 mm, Mini Protein Gels	1 box (10 gels)	NW04120BOX

* iBlot 3 iBind Flex Starter Kit includes similar components to the iBlot 3 iBind Starter Kit.

Learn more at **thermofisher.com/iblot3**

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