

# Branching out

## Exposing angiogenesis's role in cancer biology

A primary focus of cancer biology is the study of angiogenesis in tumor growth. Oxygen, nutrient availability, enzymes, and coagulating agents as well as tumor cells' ability to advance beyond the tumor are all critical components in angiogenesis.<sup>1,2</sup>

Angiogenesis is controlled by agonists and antagonists. Malignancy is marked by activating a pro-angiogenic switch that can oppose normal negative regulatory mechanisms.<sup>3</sup> Tumor staging is based on increases in tumor vascularity and pro-angiogenic factors, which in turn are predictive of poor clinical outcomes.

Current theories of tumor angiogenesis suggest that existing blood vessels recruit new vessel formation through endothelial cell migration, proliferation, and invasion and their subsequent organization into vessel structure. These hypotheses are based on the discovery of pathways for multiple growth factors, including VEGF.

VEGF and VEGF receptors are critical to normal and pathological angiogenesis. When this pathway is activated, endothelial cell survival, mitogenesis, migration, and differentiation result. Endothelial progenitor cells are mobilized from the bone marrow into the periphery. VEGF also helps modulate vessel permeability, resulting in protein deposition in the interstitium and angiogenesis promotion.

High VEGF levels are also associated with tumor progression and poor outcomes for most solid tumors, including colorectal, gastric, pancreatic, breast, prostate, and lung cancer, and melanoma. The VEGF pathway is therefore a major focus of biomarker discovery and drug development. Table 1 lists some key proteins, classified by functional biology. Invitrogen has many products that aid the ongoing clarification of the roles these proteins play in VEGF pathway participation in tumor angiogenesis.

### Invitrogen phosphoELISA™ kits

Invitrogen's phosphoELISA™ kits have also illuminated the relationship between the two VEGF receptors and cancer (Figure 1). HUVEC and mouse endothelial cells were serum starved overnight, then treated with VEGF (Cat. no. PHG0141) at 100 ng/ml for 5 minutes. Cell extracts were prepared and analyzed with the VEGFR2 [pY1059] ELISA Kit (Cat. no. KHO0601) and VEGFR2 (Total) ELISA Kit (Cat. no. KHO0591). Phosphorylation of VEGFR2 is upregulated in VEGF-treated vascular endothelial cells, while levels of total VEGFR2 remain relatively constant in VEGF-treated and untreated controls. Visit our web page [www.invitrogen.com/ELISA](http://www.invitrogen.com/ELISA) for ELISAs for angiogenesis research.

As cancer research continues to uncover the relationship between VEGF and its receptors and human cancers, Invitrogen is dedicated to providing the tools and services needed to boost this research. Visit our web page [www.invitrogen.com/proteins](http://www.invitrogen.com/proteins) for recombinant angiogenesis proteins, and [www.invitrogen.com/antibodies](http://www.invitrogen.com/antibodies) for angiogenesis-related antibodies.

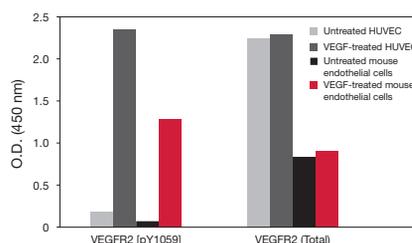


Figure 1—VEGFR2 [pY 1059] and VEGFR2 (total) ELISAs on VEGF-treated endothelial cells.

Table 1—Key VEGF regulators in tumor cells.

Oncogenes/ tumor suppressor genes	Activated signaling pathways/ transcription factors	Environmental and growth factors, cytokines, cytokine receptors
<i>Ras</i>	Src	Hypoxia
<i>Src</i>	β-catenin	Low pH
PTEN	HIF	IGF-1R
p53	COX-2	EGFR
<i>Wnt</i>	AP-1	IL-1β
Rb	SP-1	IL-6
<i>HER-2</i>	NFκB	c-Met
<i>Jun</i>	Bcl-2	PDGFR
<i>Fos</i>	Akt/PKB	
MDM2		
eIF-4E		
NOX-1		

### References

1. Folkman, J. (1990) *J Natl Cancer Inst* 82:4–6.
2. Hanahan, D., Weinberg, R.A. (2000) *Cell* 100:57–70.
3. Ellis, L., Hicklin, D. (2005) *J Clin Oncol* 23:1011–1027.



www.invitrogen.com



The most powerful cell sorter on the planet.

Introducing MoFlo™ XDP.

**X**treme Applications. **X**treme Performance. **X**treme Support.

Accelerate your cell sorting research. The power of MoFlo XDP provides zero dead time, automated droplet stream control, and the most advanced software platform and application capabilities on earth. Discover what you can achieve. Go to extremes. Go to Dako. [www.dako.com/MoFloXDP](http://www.dako.com/MoFloXDP)



*Let's connect.*

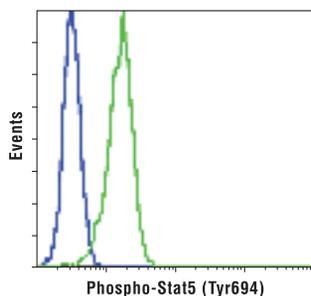


INNOVATIVE DISCOVERY TOOLS FOR SIGNAL TRANSDUCTION RESEARCH

# Over 300 Antibodies Validated for Flow Cytometry...from Cell Signaling Technology

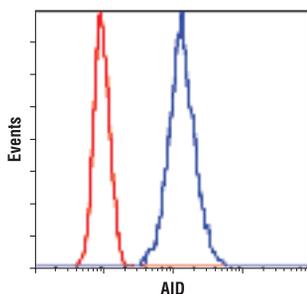


**Phospho-Stat5 (Tyr694) (C11C5)  
Rabbit mAb #9359**



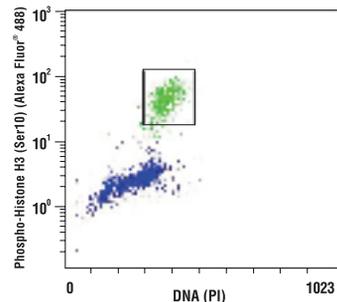
Flow cytometric analysis of K562 cells, untreated (green) or ST1571-treated (blue), using #9359.

**AID (EK2 5G9) Rat mAb #4959**



Flow cytometric analysis of Raji cells using #4959 (blue) compared to a nonspecific negative control antibody (red).

**Phospho-Histone H3 (Ser10) Antibody  
(Alexa Fluor® 488 Conjugate) #9708**



Flow cytometric analysis of THP-1 cells treated with paclitaxel using #9708 versus propidium iodide (DNA content). The box indicates phospho-histone H3 positive cells.

- ∴ Highest quality antibodies from CST provide the brightest signal with the lowest background.
- ∴ Extensive validation by our in-house clinical applications group — optimization is not left up to you, the user.
- ∴ Technical support provided by our clinical applications group — this translates into a thorough, fast and accurate response.
- ∴ Rapidly expanding line of Alexa Fluor® conjugated primary antibodies available for use in flow cytometry.

Alexa Fluor® is a registered trademark of Molecular Probes, Inc.



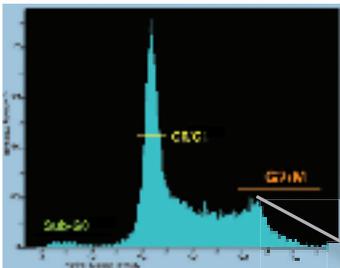
For a complete product listing please visit...

[www.cellsignal.com](http://www.cellsignal.com)

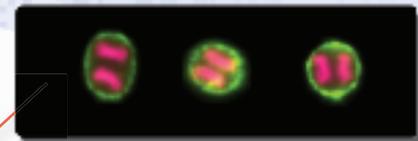
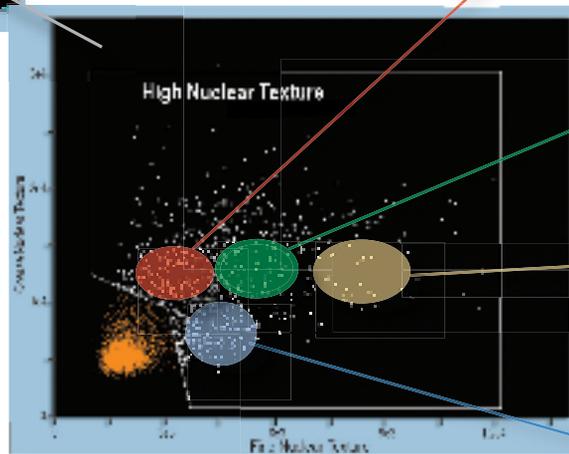
Orders (toll-free) 1-877-616-CELL (2355) [orders@cellsignal.com](mailto:orders@cellsignal.com) Fax ordering 1-978-867-2488 Technical support (toll-free) 1-877-678-TECH (8324) [support@cellsignal.com](mailto:support@cellsignal.com) Inquiries [info@cellsignal.com](mailto:info@cellsignal.com)

# In flow cytometry, every dot tells a story ...

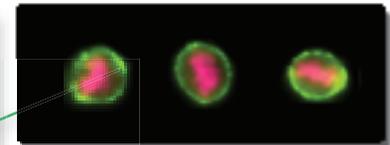
## The ImageStream® system



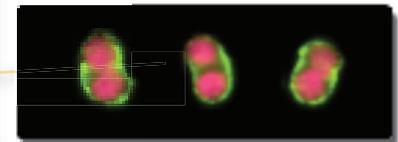
HL60 cells were labeled with AF488 anti-HLA to reveal the cell membrane and with DRAQ5™ to stain the nucleus. Labeled cells were analyzed on the ImageStream system. The DNA content histogram (above) clearly separates the major cell cycle subpopulations. G2/M cells were plotted (right) to display the high nuclear texture fraction, from which cells in the progressive stages of mitosis were identified and highlighted on the plot. Composite cell images (far right) show HLA and nuclear staining.



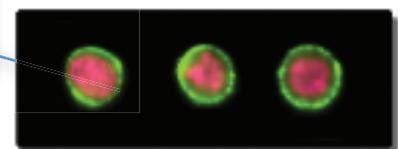
Anaphase



Metaphase



Telophase

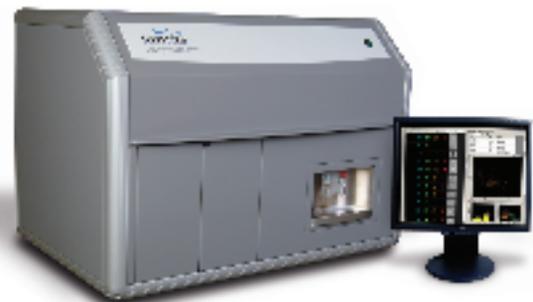


Prophase

For over 30 years, flow cytometry has been about the dots. But behind every dot is a cell, rich in morphological complexity. Now, the ImageStream imaging flow cytometer from Amnis makes this wealth of biological information directly accessible. By combining quantitative microscopy and the statistical power of flow cytometry in a single platform, the ImageStream offers you completely new options for cell analysis.

Study the intracellular distribution of markers, analyze interactions between cells, quantitate subtle morphologic changes in the cell and its structures, and much more. The range of applications is extensive, as is the growing list of published science.

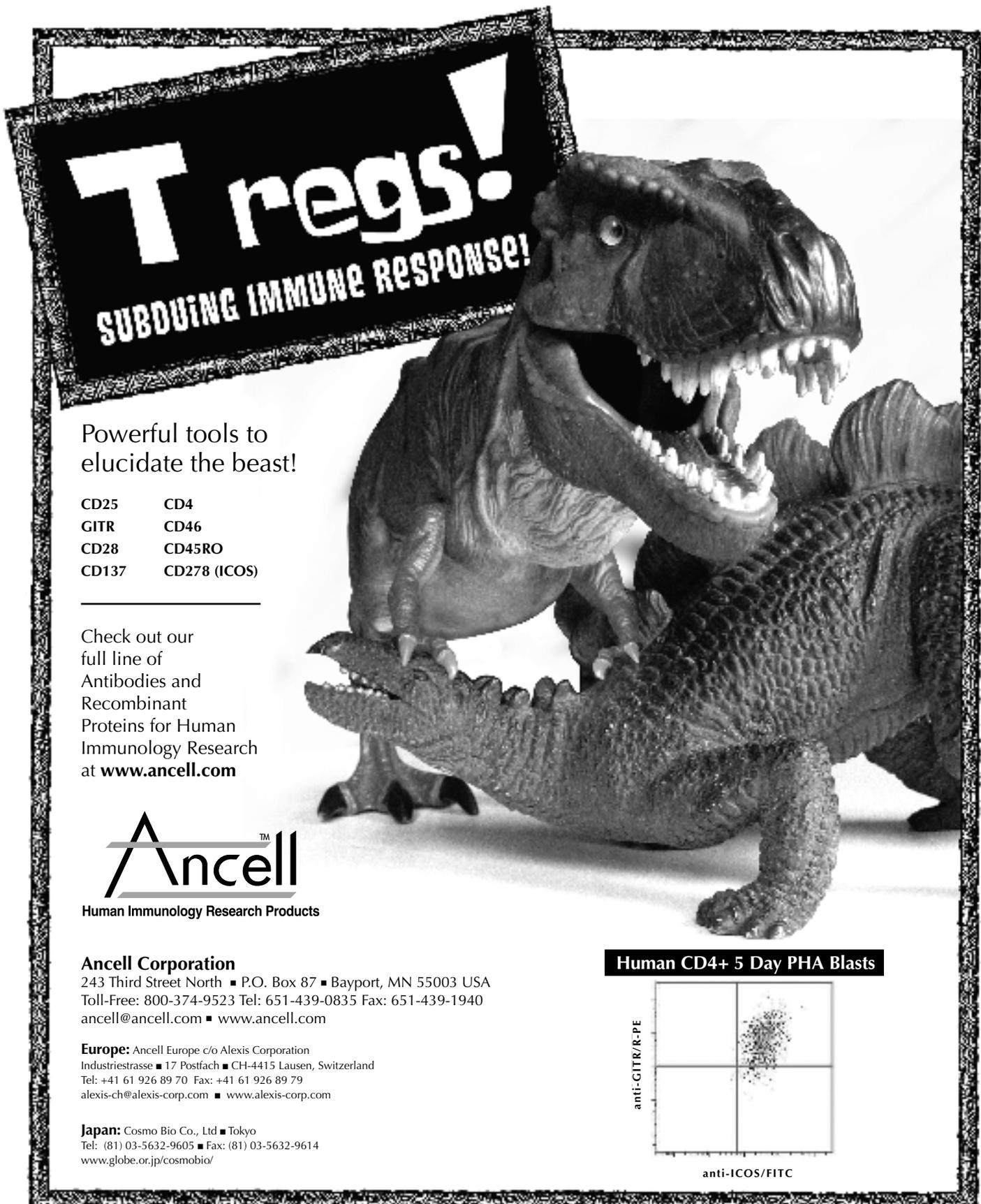
While we can't guarantee that every experiment you do with the ImageStream system will turn into a publication, we do guarantee that when you analyze your cells on the ImageStream you will see more -- and know more -- than you ever thought possible.



To learn more, visit [www.amnis.com](http://www.amnis.com) or call 800-730-7147 toll-free in the US or +1 206-374-7000 outside the US.

**ImageStream. Think outside the dot.**





**Tregs!**  
SUBDUING IMMUNE RESPONSE!

Powerful tools to elucidate the beast!

- CD25      CD4
- GITR      CD46
- CD28      CD45RO
- CD137    CD278 (ICOS)

Check out our full line of Antibodies and Recombinant Proteins for Human Immunology Research at [www.ancell.com](http://www.ancell.com)



Human Immunology Research Products

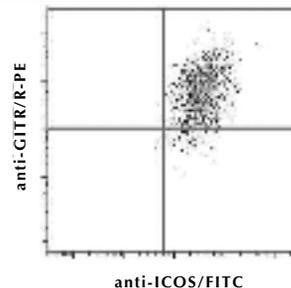
**Ancell Corporation**

243 Third Street North ■ P.O. Box 87 ■ Bayport, MN 55003 USA  
Toll-Free: 800-374-9523 Tel: 651-439-0835 Fax: 651-439-1940  
[ancell@ancell.com](mailto:ancell@ancell.com) ■ [www.ancell.com](http://www.ancell.com)

**Europe:** Ancell Europe c/o Alexis Corporation  
Industriestrasse ■ 17 Postfach ■ CH-4415 Lausen, Switzerland  
Tel: +41 61 926 89 70 Fax: +41 61 926 89 79  
[alexis-ch@alexis-corp.com](mailto:alexis-ch@alexis-corp.com) ■ [www.alexis-corp.com](http://www.alexis-corp.com)

**Japan:** Cosmo Bio Co., Ltd ■ Tokyo  
Tel: (81) 03-5632-9605 ■ Fax: (81) 03-5632-9614  
[www.globe.or.jp/cosmobio/](http://www.globe.or.jp/cosmobio/)

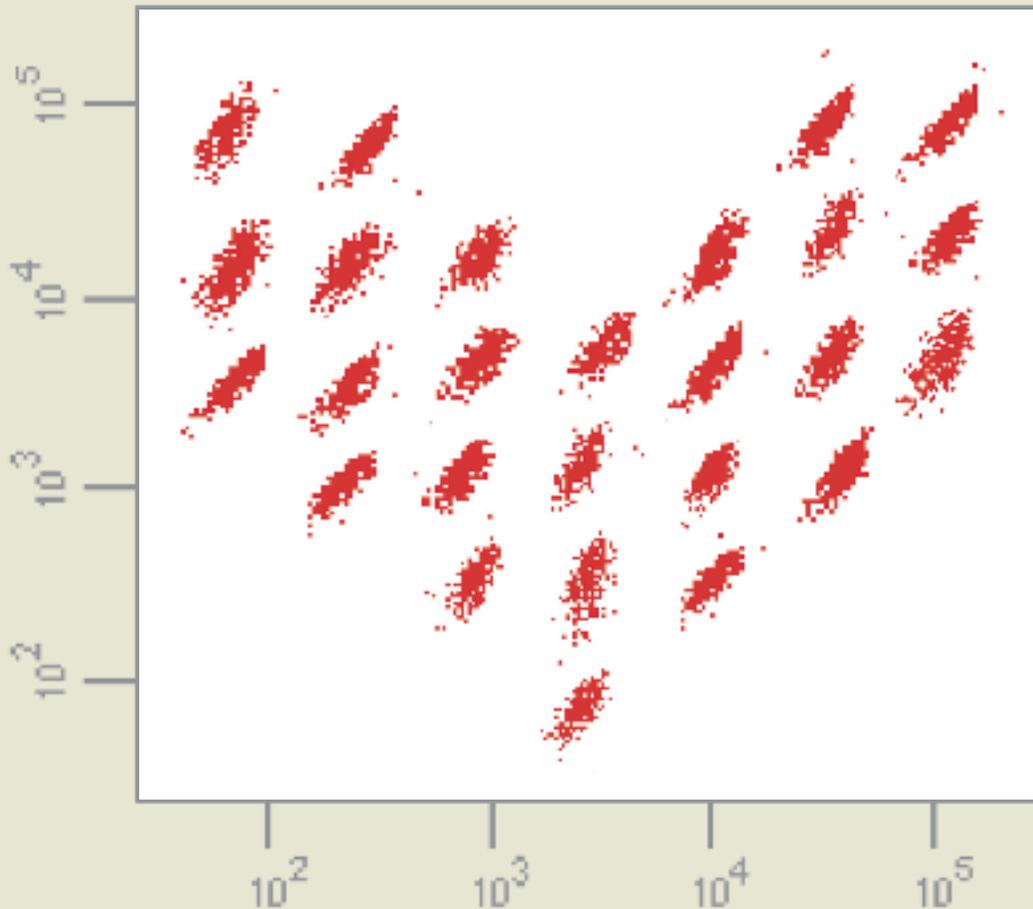
**Human CD4+ 5 Day PHA Blasts**



**International Distributors:** Austria: EUBIO (01) 895 0145 Australia: Immuno Diagnostics 612 9519 9300 Bangladesh: FUTURE BUSINESS VISION (0)2 863 1173 Belgium: 10 P's BVBA 03 466 04 20 Canada: ID LABS inc. 800-463-4782 or Fax (519) 434-2639 MEDICORP INC 514-733-1900 or Fax 514-733-1212 Czech Republic: GENETICA S.R.O. (02)7270 1055 Denmark: MEDINOVA SCIENTIFICAS 039 56 2000 Europe: ANCELL EUROPE c/o ALEXIS CORPORATION +41 61 926 89 70 Finland: NUPPULINNAN LABORATORIOPALVELU OY (09) 27940200 France: COVALAB +33 437 654 2300/ COGER S.A. (01) 45 33 67 17 Germany: AXSORA DEUTSCHLAND GmbH +49 6401 90077 Greece: BALCAN-MED (1) 8641714 SB BIOTECHNOLOGY SUPPLIERS A.E. (210) 823 3373 Hong Kong: BOPPARD (H.K.) CO., LTD 2799 9019 Hungary: BIOMARKER LTD 28 419 986 India: HYSEL INDIA Pvt. Ltd. 11 2622 78 01 Indonesia: ITS INDONESIA (021) 451 6222 Ireland: ALPHATECHNOLOGIES LTD. 045 865 440 Israel: ALMOG DIAGNOSTIC & MEDICAL EQUIPMENT LTD. (03) 977 3390 Italy: VINCI-BIOCHEM 0571 568147 Japan: COSMO BIO CO., LTD (81) 03-5632-9605 Iwai Chemicals Co., Ltd. (81) 03-3255-2781 Malaysia: INTERSCIENCE SDN BHD (03) 5740 9888 New Zealand: SAPPHIRE BIOSCIENCE PTY LTD 02 9698 2022 Norway: AH DIAGNOSTICS AS (23) 23 32 60 Pakistan: THE WORLDWIDE SCIENTIFIC. (042) 755 2355 Poland: BIOMIBO (022) 872 07 97 Portugal: BAPTISTA MARQUES, LDA (21)722 06 60 PR China: Dakewe Biotech Co. +86 255 26412280 Romania: MEDIST SA (21) 411 5003 Russia: CHIMMED INC. (095) 728 4192 Singapore: ITS SCIENCE & MEDICAL PTE. LTD. (06)273 0898 South Korea: CORNELL BIOTECH CORP. 82 2 555 0094 ResearchNET Inc. 82-2-572-5430 Spain: GRUPO TAPER S.A. (91) 484 1960 Sweden: KELAB 031 125160 Switzerland: ALEXIS CORPORATION +41 61 926 89 89 Taiwan: GenHot Laboratories Inc. 886-2-7927398 Thailand: ITS THAILAND CO. LTD. (02) 308 0611 The Netherlands: 10 P's 076 542 51 84 Turkey: BioGen 90 212 588 23 55 TOKRA MEDICAL LTD. (312) 395 60 09 UK and Republic of Ireland: AXSORA (UK) LTD. +44 (0) 1949 836111

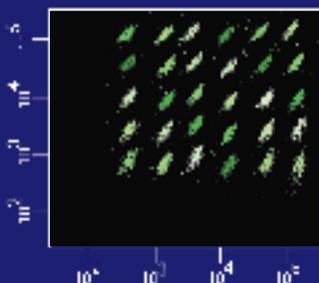
# BD™ Cytometric Bead Array

Flex Set Bead-Based Immunoassays



Helping all people  
live healthy lives

Easy to multiplex. Easy to love.



Human cytokine 30-plex acquired on  
the BD FACSAArray bioanalyzer

Stop doing time-consuming ELISA and western blot assays and save valuable samples using BD™ CBA Flex Set bead-based immunoassays for measurement of cytokines and cell signaling proteins. Measure multiple proteins simultaneously with our easy-to-use system, including pre-optimized assays, buffers, and setup reagents.

Our simple protocol requires minimal hands-on time, leaving you to concentrate on what really matters: your results.

Visit us at [bdbiosciences.com/flexset](http://bdbiosciences.com/flexset) to learn more.

Shop online at [shopbdbiosciences.com](http://shopbdbiosciences.com)

This product is for Research Use Only. Not for use in diagnostic or therapeutic procedures.  
BD, BD Logo and all other trademarks are the property of Becton, Dickinson and Company. ©2007 BD.  
06-810038-9

**BD Biosciences**  
2350 Qume Drive  
San Jose, CA 95131  
[bdbiosciences.com](http://bdbiosciences.com)