SPECIFICATION SHEET Attune flow cytometers

## Attune flow cytometer systems

Table 1. Invitrogen™ Attune™ flow cytometer specifications.

| Instrument specifications            |   | Attune™ CytPix™ Flow Cytometer  |                 | Attune™ Nx | T Flow Cytometer        |                    |
|--------------------------------------|---|---|-----------------|------------|-------------------------|--------------------|
|                                      |   | Laser   | Wavelength (nm) | Beam-shap  | oing optics (BSO)* (mW) | Diode power** (mW) |
| Optics:<br>fluorescence<br>detection | Laser power                               | Violet<br>Blue  | 405<br>488      |            | 50<br>50                | 100                |
|                                      |   | Green†  | 532             |            | 100                     | 140                |
|                                      |   | Yellow  | 561             |            | 50                      | 100                |
|                                      |   | Red   | 637             |            | 100                     | 140                |
|                                      |   | * Amount of measured usable laser power after light has gone through the beam optics and shaping filters.  ** Vendor-specified theoretical maximum.  † Green laser not available on the Attune CytPix Flow Cytometer. |                 |            |                         |                    |
|                                      | Laser excitation                          | Optimized excitation for minimized stray laser-line noise and losses to reflection  |                 |            |                         |                    |
|                                      | Laser profile                             | 10 x 50 μm flat-top laser provides robust alignment   |                 |            |                         |                    |
|                                      | Emission filters                          | Up to 14 color channels with wavelength-tuned photomultiplier tubes (PMTs); user-changeable, keyed filters  |                 |            |                         |                    |
|                                      | Laser separation                          | 100 μm  |                 | 150 µm     |                         |                    |
|                                      | Optical alignment                         | Fixed alignment with prealigned welded fiber; no user maintenance required  |                 |            |                         |                    |
|                                      | Onboard thermoelectric cooler             | No warm-up delay; fiber unaffected by "on/off"  |                 |            |                         |                    |
|                                      | Simmer mode                               | Instant on/off reduces wear up to 10-fold; only on during data acquisition; hours of usage reported   |                 |            |                         |                    |
|                                      | Flat-top laser specified at the flow cell | Coefficient of variation (CV) <3% over the width of the flat-top laser  |                 |            |                         |                    |
|                                      | Upgradable                                | Convenient field changes  |                 |            |                         |                    |
| Optics:<br>imaging                   | Laser excitation                          | 405 nm  |                 | NA         |                         |                    |
|                                      | Pulse width                               | <50 ns  |                 | NA         |                         |                    |
|                                      | Flow cell                                 | Quartz <sup>™</sup> cuvette gel coupled to 1.2 numerical aperture (NA) collection lens, 200 x 200 µm  |                 |            |                         |                    |
|                                      | Sample analysis volume                    | 20 μL to 4 mL   |                 |            |                         |                    |
|                                      | Custom sample flow rates                  | 12.5–1,000 μL/min   |                 |            |                         |                    |
|                                      | Sample delivery                           | Positive displacement syringe pump for volumetric analysis  |                 |            |                         |                    |
|                                      | Sample tubes                              | Accommodates 17 x 100 mm to 8.5 x 45 mm tubes   |                 |            |                         |                    |
| Fluidics                             | Fluid level sensing                       | Active  |                 |            |                         |                    |
|                                      | Standard fluid reservoirs                 | 1.8 L focusing fluid tank, 1.8 L waste tank, 175 mL shutdown solution tank, and 175 mL wash solution tank   |                 |            |                         |                    |
|                                      | Fluid storage                             | All fluids stored within instrument   |                 |            |                         |                    |
|                                      | Extended fluidics option                  | 10 L fluid configuration  |                 |            |                         |                    |
|                                      | Nominal fluid consumption                 | 1.8 L/day   |                 |            |                         |                    |
|                                      | Automated maintenance cycles              | ≤15 min start-up and shutdown; deep clean, sanitize, and debubble modes   |                 |            | e modes                 |                    |



Table 1. Attune flow cytometer specifications (continued).

| Instrument specifications                 |                                      | Attune CytPix Flow Cytometer  | Attune NxT Flow Cytometer   |  |  |  |
|---|--------------------------------------|---|---|--|--|--|
|   | Fluorescence sensitivity             | ≤80 molecules of equivalent soluble fluorochrome (f   | MESF) for FITC, ≤30 MESF for PE, ≤70 MESF for APC                                 |  |  |  |
|   | Fluorescence resolution              | CV <3% for the singlet peak of propidium iodide-stained chicken erythrocyte nuclei (CEN)  |   |  |  |  |
|   | Data acquisition rate                | Up to 35,000 events/sec, 34 parameters, based on a 10% coincidence rate per Poisson statistics  |   |  |  |  |
|   | Maximum electronic speed             | 65,000 events/sec with all parameters   |   |  |  |  |
|   | Carryover                            | Single tube format: <1%   |   |  |  |  |
| Performance:<br>fluorescence<br>detection | Forward and side scatter sensitivity | Can distinguish platelets from noise  |   |  |  |  |
|   | Forward and side scatter resolution  | Optimized to resolve lymphocytes, monocytes, and granulocytes in lysed whole blood  |   |  |  |  |
|   | Forward scatter                      | Photodiode detector with 488/10 nm bandpass filter  |   |  |  |  |
|   | Side scatter                         | PMT with default 488/10 nm bandpass fillter;<br>optional 405/10 + OD2 bandpass filter   | PMT with default 488/10 nm bandpass filter;<br>optional 405/10 nm bandpass filter |  |  |  |
|   | Fluorescence detectors               | 14 individual detectors   |   |  |  |  |
|   | Electronic pulse                     | Measured area; height and width pulse for all detectors   |   |  |  |  |
|   | Violet side scatter resolution       | Can be configured for violet side scatter to better resolve particles from noise  |   |  |  |  |
|   | Minimum particle size                | 0.2 µm on side scatter using submicron bead calibration kit from Bangs Laboratories or 0.1 µm on side scatter under followin conditions: use an Attune NxT Flow Cytometer with standard 0.5 mm blocking configuration, an Invitrogen™ Attune™ NxT 488/Filter (Cat. No. 100083194), and Invitrogen™ Attune™ Focusing Fluid (Cat. No. 4488621, 4449791, or A24904) that has been passed through a 0.025 µm filter |   |  |  |  |
|   | Pixel resolution                     | 0.3 µm/pixel  | NA  |  |  |  |
|   | Objective magnification              | 20x   | NA  |  |  |  |
|   | Objective numerical aperture (NA)    | 0.45  | NA  |  |  |  |
| erformance:                               | Theoretical resolution               | 0.6 µm  | NA  |  |  |  |
| maging                                    | Detection limit                      | Visually detect 800 nm particles  | NA  |  |  |  |
|   | Image capture rate                   | Up to 6,000 images/second depending on image size and event rate  | NA  |  |  |  |
|   | Image size                           | 96 x 96 pixels to 248 x 248 pixels  | NA  |  |  |  |
|   | Field of view                        | 29 x 29 μm² to 74 x 74 μm²  | NA  |  |  |  |
|   | Compensation                         | Full matrix in automated and manual modes; on-plot compensation tools for fine adjustment; use of tubes and wells   |   |  |  |  |
|   | Flow rate                            | Precise flow rate control via software; no hardware adjustments   |   |  |  |  |
|   | Live streaming                       | Live update of statistics during event acquisition up to 35,000 events/sec  |   |  |  |  |
|   | Overlays                             | Comparative analysis between samples; 3D view   |   |  |  |  |
|   | Sample recovery                      | System able to return unused samples  |   |  |  |  |
|   | Concentration                        | Direct concentration measurement without use of counting beads  |   |  |  |  |
|   | Software layout                      | Fully customizable for each user account  |   |  |  |  |
|   | Bubble detection technology          | Stops automated run to preserve sample integrity  |   |  |  |  |
|   | Maximum single-event file            | 20 million with option to append  |   |  |  |  |
| low                                       | Heat map                             | Set up for definition of plate layout; screening view for analysis for tubes and plates   |   |  |  |  |
| cytometry<br>software                     | Threshold                            | Up to 4 individual thresholds with user option to apply Boolean logic   |   |  |  |  |
| eatures                                   | Gating                               | Hierarchical gating with the ability to derive gates  |   |  |  |  |
|   | Voltage                              | User adjustable   |   |  |  |  |
|   | Window extensions                    | User adjustable   |   |  |  |  |
|   | Area scaling factor (ASF)            | User adjustable   |   |  |  |  |
|   | Acquisition settings                 | Documented in FCS files and maintained upon import  |   |  |  |  |
|   | Templates                            | Create from existing experiments—instrument settings, workspaces, run protocols, heat map settings, and compensation settings optimized and defined previously  |   |  |  |  |
|   | Tube-to-plate conversion             | One-click transition from tubes to plates and vice versa; no disassembly, no additional QC, no reboot required for conversion between plates and tubes  |   |  |  |  |
|   | Graphics resolution                  | Publication-quality data plots; supports TIF, PNG, BMP, JPG, GIF, and EMF files;<br>quickly copy and paste plots to any external application (e.g., Microsoft™ PowerPoint™ software)  |   |  |  |  |
|   | User account                         | Administrative creation of individual user accounts with designated roles; advanced setting permissions; management of individual accounts; user time tracking and sample count   |   |  |  |  |

Table 1. Attune flow cytometer specifications (continued).

| Instrument specifications (continued) |                                    | Attune CytPix Flow Cytometer  | Attune NxT Flow Cytometer  |  |  |  |
|---------------------------------------|------------------------------------|---|--|--|--|--|
|                                       | Image capture settings             | Set total number of recorded images, image frequency, image capture gate, image size, image position, focus, and illumination for control over experiment design and data footprint.  | NA   |  |  |  |
| Imaging<br>software<br>features       | Image view                         | Image view option allows overview of image gallery with cell image option to view individual images in the workspace for any cell population.   | NA   |  |  |  |
|                                       | Image backgating                   | Correlate images to flow cytometry data by backgating all or only selected images onto supported workspace plots.   | NA   |  |  |  |
|                                       | Image measurement tool             | Elliptical tool to measure event areas in images in µm²   | NA   |  |  |  |
|                                       | Image export options               | Exports images as 8-bit TIF, PNG, GIF, BMP, JPG, or EMF files   | NA   |  |  |  |
| Quality and regulatory                | Instrument tracking                | Automated daily baseline and performance test with Levey-Jennings plots   |  |  |  |  |
|                                       | Warranty                           | 1 year  |  |  |  |  |
|                                       | Production verification testing    | Each instrument is tested and verified for assembly integrity and performance to specifications   |  |  |  |  |
|                                       | Quality management system          | Manufacturing standards comply with the requirements of ISO 13485:2003  |  |  |  |  |
|                                       | Robust installation specifications | Units installed by engineer; preplanning checklist, delivery, and installation; and performance validation compliance with standardized procedure   |  |  |  |  |
|                                       | Regulatory status                  | For Research Use Only   |  |  |  |  |
|                                       | Software requirements              | Invitrogen™ Attune™ Cytometer Software  |  |  |  |  |
|                                       | Monitor                            | 27 in. flat panel (1,920 x 1,080 resolution);<br>dual-monitor capability  | 23 in. flat panel (1,920 x 1,080 resolution);<br>dual-monitor capability         |  |  |  |
|                                       | Computer                           | Minitower desktop   |  |  |  |  |
|                                       | Operating system                   | Microsoft™ Windows™ 10 software, 64-bit   |  |  |  |  |
| Computer                              | FCS format                         | FCS 3.1, 3.0  |  |  |  |  |
|                                       | Processor                          | Intel <sup>™</sup> Core <sup>™</sup> i7 processor   |  |  |  |  |
|                                       | RAM                                | 64 GB   | 32 GB  |  |  |  |
|                                       | Hard drives                        | 2 x 8 TB SSD, 560 MB/sec;<br>controller RAID1, integrated   | 2 x 2 TB SATA 3.0 Gb/sec, 8 MB data burst cache;<br>controller RAID1, integrated |  |  |  |
|                                       | GPU                                | NVIDIA™ Quadro™ P2200   | NA   |  |  |  |
| Installation requirements             | Electrical requirements            | 100-240 VAC, 50/60 Hz, <150 W  Thermo Fisher Scientific certifies that the Attune flow cytometers conform to relevant directives to bear the CE mark.  The instrument also conforms to the UL and CAN/CSA general requirements (61010.1). The Attune flow cytometers are class I laser products per Center for Devices and Radiological Health (CDRH) regulations and EN/IEC 60825. |  |  |  |  |
|                                       | Heat dissipation                   | <150 W  |  |  |  |  |
|                                       | Temperature operating ranges       | 15–30°C (59–86°F)   |  |  |  |  |
|                                       | Operating humidity                 | 10-80%, noncondensing   | 10-90%, noncondensing  |  |  |  |
|                                       | Audible noise                      | <65 dBA at 1.0 m  |  |  |  |  |
|                                       | Instrument size<br>(H x W x D)     | ~49 x 58 x 43 cm (19 x 23 x 17 in.), including fluid bottles  | $\sim$ 40 x 58 x 43 cm (16 x 23 x 17 in.), including fluid bottles               |  |  |  |
|                                       | Weight                             | ~33 kg (73 lb)  | ~29 kg (64 lb)   |  |  |  |

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## Autosamplers for Attune flow cytometers

Table 2. Technical specifications.

| Specifications                            | Invitrogen™ CytKick™ Autosampler   | Invitrogen™ CytKick™ Max Autosampler   |  |  |
|---|--|--|--|--|
|   | <ul> <li>&lt;42 min per 96-well plate in high-throughput mode</li> <li>&lt;70 min per 96-well plate in standard mode with wash cycles</li> </ul> | • 22 min per 96-well plate in Boost mode; one rinse, one mix, and full analysis for each 20 µL sample at 1,000 µL/min  |  |  |
| Acquisition time                          | • <145 min per 384-well plate in standard mode; one mix, one rinse, and full analysis for each 20 µL sample at 500 µL/min                        | • 88 min per 384-well plate in Boost mode; one rinse, one mix, and full analysis for each 20 µL sample at 1,000 µL/min |  |  |
|   | • <0.5% carryover for 100 µL, 200 µL, 500 µL, and 1,000 µL samples with one mix and one rinse in standard mode                                   | • <0.5% carryover for 100 µL, 200 µL, 500 µL, and 1,000 µL samples with one mix and one rinse in standard mode         |  |  |
| Carryover                                 | • <1.0% carryover for 12.5 µL and 25 µL samples  | <ul> <li>&lt;1.0% carryover for 12.5 μL and 25 μL samples</li> </ul>   |  |  |
|   |  | • <1.0% carryover for 500 µL and 1,000 µL samples in Boost mode with one mix and one rinse                             |  |  |
| Mixing optimization                       | Mixing optimized to preserve cell viability; number of n   | nixing cycles optimized to match sample analysis volume  |  |  |
| Mixing method                             | Each well mixed via aspiration and dispensation of sample (no shaking)   |  |  |  |
| lo. of wash cycles                        | Up to 10 wash cycles (user-defined)  |  |  |  |
| Minimum dead volume (single draw)         | 30 µL for 12.5–200 µL/min; 50 µL for 1,000 µL/min  |  |  |  |
| Sample window                             | Window allows viewing of well progress; protective coating prevents exposure to ambient light during acquisi                                     |  |  |  |
| Autocalibration                           | Regular 30-day intervals with system-initiated function  |  |  |  |
| Plate and tube compatibility              | •  | versa; no disassembly, no additional QC, no reboot   |  |  |
|   | 96 deep-well (flat, U-bottom, and V-bottom)  | 96 deep-well (flat, U-bottom, and V-bottom)  |  |  |
|   | 96-well standard depth (flat, U-bottom, and V-bottom)  | 96-well standard depth (flat, U-bottom, and V-bottom)  |  |  |
|   | 384-well standard depth (flat, U-bottom, and V-bottom)   | 384-well standard depth (flat, U-bottom, and V-bottom)   |  |  |
|   | 384 deep-well (flat, U-bottom, and V-bottom)   | 384 deep-well (flat, U-bottom, and V-bottom)   |  |  |
| Compatible plate types                    |  | Customizable to accept other plate types   |  |  |
|   |  | 1.5 mL and 2 mL microcentrifuge tube rack<br>(up to 24 tube racks per vessel)  |  |  |
|   |  | Foil-covered 96-well (U-bottom) and 384-well<br>(U-bottom and V-bottom)  |  |  |
| Fluidics requirements                     | Fluid storage: external Total fluid volume: two 2 L tanks  |  |  |  |
| Extended fluidics                         |  | (EFS); optional external fluid tank with 10 L fluid capacity   |  |  |
| size (W x D x H)                          | ~43 x 33 x 41 cm (17 x 13 x 16 in.)  |  |  |  |
|   | Minimum width: 43 cm (17 in.); total width is 99 cm (39 in.) when attached to an Attune flow cytometer   |  |  |  |
| Space requirements                        | • Minimum depth: 39.5 cm (15.6 in.); allow 33 cm (13 in.) for the cytometer unit and 6.5 cm (2.6 in.) behind the unit for ventilation            |  |  |  |
|   | Minimum clearance height: 74 cm (29.1 in.) above the mounting  |  |  |  |
| Mounting                                  | Mounted on side or placed behind   |  |  |  |
|   | 16.9 kg (37.2 lb) with empty focus and waste bottles   |  |  |  |
| Weight                                    | 20.9 kg (46 lb) with focus and waste bottles at full capacity  |  |  |  |
| Operating range environmental conditions) | 20.9 kg (46 lb) with focus and waste bottles at full capacity  15–30°C (59–86°F)   |  |  |  |
| Operating humidity                        | <80%, noncondensing  |  |  |  |
| Electrical requirements                   | <80%, noncondensing<br>100–240 VAC, 50/60 Hz, <300 W   |  |  |  |
| Sample cooling                            | NA   | Passive cooling available for 96-well U-bottom plates and microcentrifuge tube racks                                   |  |  |
| Evaporation protection (foil cover)       | NA   | Yes  |  |  |
| Service                                   | Field service or ra  | pid exchange option  |  |  |
|   | 1-year standard warranty; extended warranty options available  |  |  |  |

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