Table 2. Manipulation techniques: on-chip sorting of *C. elegans*.

Sorting technique	Principle	*C. elegans* population	Selectivity	Throughput	Manner	Chip arrangement / structure	Technology	Application	Picture	References
Electrotaxis	DC (3.5–7 V/cm); AC (20 mHz–1 Hz); DC pulse (50 µs, 1 Hz–1 kHz, 3 V/cm–12 V/cm)	Worm population (L2–L4 stage larvae, adults, and mutants)	Middle (70–90 %)	Low (4.3±1.0 worms/min) and high (78 worms/min per load run)	Passive, long-term (~40–60 min)	Line-channels with embedded electrodes in inlet and outlet reservoirs	Single PDMS layer	Drug screening, age- and size synchronization, movement-based behavioural studies	Fig. 2A	30-33, 38, 52 and 58
"On-flow" sorting	Self-regulated sample loading	Worm population (larvae and adults)	High (94% of adults with 0.2% larva contamination)	High (200 worms/min)	Passive, long-term	Microstructered architecture "smart mazes" or microchambers	Single PDMS layer	Age- and size synchronization, culturing, stimulation	Fig. 3	21, 37, 45, 50, 55, 68, 101, 102 and 105
"On-valve" sorting	On/off valve switching	Single worm	High (96.5%)	Low (50 worms/h) and high (400 worms/h)	Active, long-term	Two outlets , positioning area, detection area	Single, two, and multi PDMS layers	Stimulation, phenotyping, microsurgery	Fig. 6C	28, 47, 51, 53, 56, 104 and 109
Drop encapsulation	Worm encapsulation in a drop	Single worm (synchronized L1 stage larvae)	Low (60 % in ~10 min)	High	Active, long-term (~120 min)	T-junction droplet generator and droplet trap array	Single PDMS layer	Drug screening, movement-based behavioural studies	Fig. 4	23 and 49

Table 3. Manipulation techniques: on-chip immobilization of *C. elegans*.

Immobilization technique	Principle	*C. elegans* population	Selectivity	Throughput	Manner	Chip arrangement / structure	Technology	Features	Application	Picture	References
Mechanical force	Microchannel narrowing	Single worm (adults)	High	High (400 worm/h)	Active, long-term	Two fluid outlets, positioning area, and detection area	Single, two, and multi PDMS layers	Automatic, repeatable	Movement-based behavioural studies (ageing), drug screening	Fig. 2B, Fig. 6B, Fig. 6C	11, 13, 14, 16, 17, 20, 22, 24, 28, 32, 35, 36, 37, 46, 47, 51, 56, 59, 60, 63, 64, 67, 68, 94, 95, 98, 102 104 and 110
	Compression (25 psi)	Single worm (L4 stage larvae and adults)	High	High (few s)	Active, short-term (~minutes)	Thin deformable membrane	Two and multi PDMS layers	Semi- and total immobilization	Stimulation		
	Suction flow (10–20 psi)	Single worm (L4 stage larvae, adults, and mutants)	High	High	Active, short-term (~ 5 min)	Vacuum-assisted restaraint	Single and multi PDMS layers	Automatic, repeatable	Phenotyping		
	Combinations	Single worm (adults)	High	High	Active, long-term	Vacuum-assisted restaraint with either deformable membrane or microchannel narrowing	Two and multi PDMS layers	Extremely stable	Microsurgery		
Gel	Thermo-sensitive polymer (temperature change of ~2 °C)	Worm population (L1–L4 stage larvae and adults)	High	Low (~45–60 s gel transition time)	Active, long-term	Culturing microfluidic chambers with additional control layer	Two PDMS layers	No physiological influence, minimal physical deformation, no effect on image quality, automatic, repeatable	Culturing, Movement-based behavioural studies		43, 47 and 96
Temperature	Temperature decrease (~4 °C) or increase (~37 °C)	Single worm	High	High (400 worms/h)	Active, short-term (~2 s)	Additional control layer above fluid flow channel	Two PDMS layers	Automatic, repeatable, worm viability of 100%	Phenotyping		28, 53, 68, 93 and 109
Gas	CO_2 and N_2 supply through thin membrane (~10 psi)	Single worm (L4 stage larvae and adults)	High	Low (1–2 min/cycle	Active, short-term (~1 min), and long-term (1 hour)	Additional layer with closed-end above fluid flow channel	Two PDMS layers	Total (CO_2) and partial (N_2) immobilization, automatic	Movement-based behavioural studies	Fig. 2A	22, 35 and 101
Electrotaxis	DC (18 V/cm); AC (10 Hz–3 kHz, 3–10 V/cm)	Worm population (L2–L4 stage larvae, adults, and mutants)	High (> 80%)	High	Passive, long-term	Line-channels with embedded electrodes in inlet and outlet reservoirs	Single PDMS layer	Partial immobilization, localization, not repeatable	Movement-based behavioural studies, neuronal study	Fig. 2A	29-32, 46, 75, 93 and 108
Acoustic waves	Surface acoustic wave	Worm population (adults)	High	High	Passive, short-term	Microfluidic chip with resonance band of chirped interdigital transducers	Single PDMS layer	Immobilization, possible physiological damage	Movement-based behavioural studies		62

Table 4. On-chip imaging techniques for *C. elegans*.

Imaging technique	Principle	Field-of-view	Resolution	Output	Image acquisition / throughput	Detector / illumination	Technology	Features	Drawbacks	Application	Picture	References
Fluorescence microscopy imaging	Optical imaging system	Narrow	High (pixel accuracy >99.9 %)	Colour image	Fast	CCD camera, optical system, and filters	Assembling	Age-independant, repeatable	Fluorescent labeling, photodamage, bulky, expensive	Genetic screening, sorting		7, 11, 14-17, 22, 24, 25, 28, 34-37, 41, 44, 46, 47, 53, 54, 56-58, 60, 63, 66, 68, 93, 94, 95, 96, 98, 101, 102, 105, 107, 108 and 110
Non-fluorescence microscopy imaging	Optical imaging system	Narrow	High	Colour image	Fast	CCD camera and optical system	Assembling	Label free, age-independant, repeatable	Bulky, expensive, limited information about sample	Behavioural phenotyping, monitoring		7-10, 13, 17, 18, 21-23, 25, 26, 29, 30, 31-35, 38, 39, 42, 43, 45, 47, 49, 50-52, 55, 62, 95, 96, 101, 102, 105 and 107
Contact optofluidic	Lensless	Wide	High (0.9 µm)	Colour image	High (40 worms/min)	CCD sensor, coherent light source, and aperture	Single PDMS layer on imaging sensor	Portable, low power consumption	Dependence on sample orientation	Monitoring	Fig. 5A	70 and 71
Shadow imaging	Lensless	Wide (3.2 mm x 2.5 mm)	Low (>10 µm)	Black-and-white image	Fast	CMOS sensor, coherent light source, and metal grid	Polycarbonate microchambers with polyester film and grid	Portable, low power consumption	Age-dependant, mechanical allignment	Monitoring	Fig. 5A	69
Digital holography	Lensless	Wide (>24 mm)	High (2.2 µm)	Colour and monochrome	Slow (1–4 s)	CMOS sensor, spatially incoherent light, and aperture	Sample plane on printed circuit board (PCB)	Low power consumption	Age-dependant, no real-time moniring	Monitoring, neuronal study	Fig. 5A	72-75
Micro-electrofluidic grids	Electrical resistance change of grid`s intersection regions	24 x 24 grids	30 µm x 30 µm	Pseudo colour,grayscale and binary image	High (174 Hz readout)	Two layers of microelectrode arrays	Standard photolithography	Portable, cost-effective, low power consumption	Age-dependant, not repeatable	Drug screening	Fig. 5B	40