Microfluidic Technologies for Yeast Replicative Lifespan Studies

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ABSTRACT. The budding yeast *Saccharomyces cerevisiae* has been used as a model organism for the study of aging for over 50 years. In this time, the canonical aging experiment—replicative lifespan analysis by manual microdissection—has remained essentially unchanged. Recently, microfluidic technologies have been developed that may be able to substitute for this time- and labor-intensive procedure. These technologies also allow cell physiology to be observed throughout the entire lifetime. Here, we review these devices, novel observations they have made possible, and some of the current system limitations.

Keywords

yeast, microfluidics, microscopy, replicative aging, microdissection, lifespan

1. Introduction

Although budding yeast colonies can be propagated indefinitely, in 1959, Mortimer and Johnson reported that individual yeast cells are not immortal. *S. cerevisiae* cells reproduce clonally but asymmetrically, with a smaller daughter cell budding from its larger mother¹. Mortimer and Johnson closely monitored individual mother cells grown on an agar pad and periodically used a micromanipulator to manually remove mature daughter buds. This removal of daughter cells is necessary because a single mother cell becomes indiscernible from its exponentially dividing progeny after only a handful of division cycles. In this seminal study, they reported that an individual mother cell can divide only a finite number of times—its replicative lifespan.

Since then, multiple conserved longevity pathways have been discovered in the budding yeast². With a short generation time, genetic tractability, and minimal husbandry costs, the budding yeast has become a premier model organism for aging research³. Mortimer and Johnson's microdissection technique, developed over fifty years ago, remains the bread-and-butter yeast aging assay⁴. Unfortunately, it is also time and labor intensive. As a consequence, many labs forgo replicative lifespan studies, and, even among those that perform them, sample sizes are often too small for rigorous statistical confidence in published results. For example, the only genome-wide analysis of replicative lifespan performed to date utilized an iterative strategy involving lifespan analysis of only 5 cells per deletion strain^{5,6}. Thus, in many ways, the standard replicative lifespan assay has become a rate-limiting step in the progress of yeast aging research.

Recently, a number of groups⁷⁻¹⁴ have developed microfluidic technologies to address this problem (**Table 1**). Microfluidic devices refer to a diverse class of objects featuring sub-millimeter chambers and channels. At this scale, fluids have interesting mechanical properties—such as laminar flow—which allow researchers to precisely manipulate tiny volumes of liquid for applications including bioassay integration, high throughput screening, and chemical synthesis among many others^{15,16}. Devices constructed from polydimethylsiloxane (PDMS)—a clear, gas-permeable, and biocompatible elastopolymer—have proven particularly useful for cell biology. Tiny cell culture chambers with precisely controlled cellular microenvironments can be created by sealing PDMS blocks with micron-sized surface features onto glass cover slips. Although an exhaustive discussion of the fabrication of such devices and their applications is beyond the scope of this article, we refer the interested reader to several recent reviews¹⁷⁻²⁰.

A wide variety of microfluidic devices, including a commercial platform, have been developed for yeast culture and investigation^{8-14,21-30}. Most, however, are only suitable for experiments lasting a few hours or less²¹⁻²⁷. Though earlier attempts were made to apply microfluidics to yeast aging research^{7,30,31}, the first device to successfully demonstrate the ability to perform a replicative lifespan analysis was published in 2012⁸. Since then, six additional microfluidic designs with lifespan analysis capabilities have been developed⁹⁻¹⁴. The devices developed for yeast aging feature clever geometrical structures to mechanically trap mother cells. These structures exploit the size difference between mother cells and newborn daughter cells or the budding patterns of mother cells, generally allowing fluid flow to selectively remove mature daughters. The effective removal of undesired cells is crucial to prevent the device from becoming overgrown with multiplying progeny. When coupled with time-lapse microscopy, these devices allow both measurement of replicative lifespans and observation of physiological changes as the trapped cells age. A typical lifespan experiment using a microfluidic device at standard 30°C

conditions lasts 2 – 5 days. In contrast, microdissection experiments performed using standard methods, where cells are incubated at 4°C overnight, can last up to 3 weeks. This mini-review will discuss these seven microfluidic devices for the whole-lifespan trapping and monitoring of budding yeast cells.

2. Device Designs

2.1 Chemical and Mechanical trapping

The first microfluidic device developed to perform a replicative lifespan experiment was published by Xie and colleagues in 2012⁸. This device uses both mechanical constraints and biotin-avidin affinity (**Figure 1A**). In this device, the height of the trapping chamber is 4 μm , approximately the diameter of a yeast cell. Additionally, the glass surface is functionalized with biotinylated bovine serum albumin followed by neutravidin. Prior to loading into the device, cells are labeled with biotin. Because daughter cell walls are generated anew, only the mother cells retain this surface mark. After loading, mother cells are immobilized both mechanically—squeezed between the PDMS ceiling and the glass floor of the chamber—and chemically—glued to the glass floor by the interaction of the cell's biotin and the device's avidin. Fluid flow preferentially removes the smaller, biotin-free daughter cells. Multiple trapping chambers are arrayed like rungs in a ladder between two taller main flow channels, which allows for chambers exposed to a range of flow speeds. Yeast replicative lifespan curves were successfully generated using this device; however, those lifespans were shorter (see **Table 2**) than the results seen from microdissection experiments on agar plates⁸.

2.2 Vertical mechanical trapping by pensile columns/micropads

Later in 2012, two groups published very similar device designs using purely geometric measures to selectively trap mother cells^{9,10}. In these designs, the trapping chamber does not have a uniform height. “Micropads” (30 by 15 μm , Lee et al.) or “pensile columns” (40 by 40 μm , Zhang et al.) descend from the PDMS ceiling to create areas where the floor to ceiling distance is only 4-5 μm (**Figure 1B**). Between the micropads/columns, the trapping chamber is much taller (15 μm for Lee et al., unspecified for Zhang et al.) Cells are loaded at high pressure, pushing the column areas upwards temporarily and allowing cells to flow underneath. When this pressure is released, the cells are trapped, sandwiched under the micropad above. Again, mother cells are preferentially retained over the smaller daughters because of the size difference. Moreover, the limited area of the micropad/column prevents overgrowth, as progeny that grow into the non-micropad areas are no longer vertically constrained and more easily washed out. However, Lee et al. reported that over 70% of cells were lost before the end of their experiments. Retention data was not reported by Zhang et al.

2.3 Trapping in elongated cavities

The CLiC (Cell Loaded into a Cavity) device features “trapping areas” (90 by 40 μm , 3.3 μm height) arranged in a grid and separated by media flush channels (40 μm height)¹¹. Each trapping area features two elongated “cavities” or dead-end hallways that run off of the main trapping area (6 or 10 by ~ 32 μm) (**Figure 1C**). This design requires a significantly larger footprint per trapped mother cell than the other devices. This may become an issue in experiments that require a large number of cells (e.g. investigating rare events) when the speed of the microscope components becomes a limiting factor. Cells are loaded through the media flush channels such that a handful are caught within each trapping area. Subsequent

cell division results in a confluent monolayer of cells in the trapping area that eventually deposits a new virgin cell (never having previously produced a daughter cell) at the dead-end tip of the elongated cavity. This method has the advantage that cells are guaranteed to be virgin at the beginning of the lifespan experiment, while the previous three designs are unable to control this ¹¹. In a log-phase culture, one would expect about half of the cells to be virgin daughters and less than 1/16 of the cells to have budded more than three times. Staining of bud scars showed that the average age of a trapped cell under a pensile column in Zhang et al. is less than two divisions at the start of an experiment ¹⁰.

The CLiC device depends on the unipolar axial budding pattern of haploid yeast cells in the BY strain background ¹¹. It is worth noting that this device, along with all devices described in this review have only been demonstrated for lifespan analyses of haploid cells and may be unsuitable for larger diploid cells, as discussed further below. If a cell at the dead-end tip of the cavity buds towards the exit, it will continue to bud in the same direction and tend to remain trapped in the cavity for its entire replicative lifespan. The limited width of the cavity ensures that daughters and further progeny are continuously pushed out of the cavity. Although cells can rotate and polarity switches occur occasionally, the authors reported retention of ~65% of trapped mother cells for their entire replicative lifespans ¹¹.

2.4 Trapping between two columns

The ALCATRAS (A Long-term Culturing and TRApping System) device utilizes traps each composed of two floor-to-ceiling columns arranged with a “\ /” shaped footprint (**Figure 1D**). When a cell suspension is flowed into the device, individual cells are caught between the two columns. Continuous, unidirectional fluid flow keeps each mother cell immobilized against the columns. The limited height of the device chamber discourages vertical stacking of cells. Like the designs described above, this device exploits the size difference between mother and daughter cells. Daughter buds are forced downstream through the gap between the two columns, detaching when mature. Buds that emerge upstream are similarly removed by fluid flow upon maturity, and the authors reported whole-lifespan retention of ~40-60% of trapped mother cells. The ALCATRAS device also does not guarantee that mothers begin the experiment as virgins. However, unlike all the previously described devices, which have non-uniform internal heights, the mold for this device requires features of only a single height.

The HYAA-Chip (High-throughput Yeast Aging Analysis) follows essentially the same principle as the ALCATRAS device, sharing the advantages of single-layer fabrication and small trap footprint. However, the cross-section of the floor-to-ceiling columns is different, as is the spacing between columns in a single trap and between traps (**Figure 1E**). The general layout and dimensions of the device are also different, and it supports up to 16 strains in parallel. These changes resulted in reports of >90% retention of wild-type mother cells for their entire lifespans ¹³. This device and trapping structure has been validated on a number of both long- and short-lived mutants (Table 1). The trapping mechanism used in both of these papers allows a higher density of cells to be imaged within a single field of view when compared with the previous designs.

2.5 Trapping in a three-column jail

The Yeast Replicator device also uses vertical floor-to-ceiling columns to trap individual cells, modifying an earlier three-column “jail” design ^{14,30} (**Figure 1F**). The three columns, arrayed at the vertices of an equilateral triangle, create a fully constrained trapping area for a single cell. When cells are loaded into the device, they are first caught between two columns outside the jail. These temporarily immobilized

cells can then bud into the central trapping area. When mature, the buds become the trapped mother cells. Like the CLiC device, the Yeast Replicator guarantees that trapped cells start in the device as virgins. Daughter buds of these trapped cells emerge in between the three columns of the jail and are washed away when mature. Retention rates were not reported by the authors¹⁴.

3. Discoveries

Coupled with time-lapse microscopy, these devices allow researchers to more easily collect replicative lifespan data. Moreover, when combined with fluorescently-labeled proteins or dyes, researchers can track changes in cell physiology (e.g. organelle structure or function^{9,11,32,33}, protein level or localization^{12,13,34,35}, promoter activity^{8,10,14}, daughter-cell birth characteristics^{36,37}) across the aging process. Importantly, these measures can be associated at single-cell resolution with lifespan or other age-associated changes (e.g. cell cycle duration). A number of interesting observations have followed. For example, researchers found that delays in nuclear segregation during cytokinesis occur rarely during a cell's lifetime but increase dramatically in frequency after division speeds slow with age³². Researchers also correlated heterogeneity in end-state cell morphology, which had been previously reported by microdissection studies^{38,39}, with molecular properties of those cells. Cells trapped under a PDMS column that die in a round (as opposed to elongated) morphology are shorter-lived, display more pronounced measures of mitochondrial dysfunction, and have higher Hsp104 promoter activity at death^{9,10}. This suggests that each yeast cell may experience one of multiple modes of cellular death. Interestingly, another report using paired-column traps correlated lifespan with the morphology of the attached daughter bud at cell death (round, elongated, or no bud) but made no mention of elongated mothers¹³. This suggests that the geometry of the trapping structure may affect trends in aged cell morphology.

Building on the widely mentioned observation that cell cycle durations increase dramatically during the last few cell divisions^{8-11,14,40}, Xie *et al.* found that mutants defective in telomerase function display stochastic episodes of cell-cycle lengthening in young cells. These strains are short-lived, display aged mitochondrial morphology, and activate more frequent bursts of DNA damage response throughout life. Interestingly, lifespan shortening is unrelated to telomere length³⁷. Xu *et al.* made similar observations when tracking cellular lineages instead of mother cells. They observed a subpopulation of lineages that experienced stochastically (but not progressively) elongated cell divisions, linking this behavior to Pol32 DNA damage repair activity⁴¹.

Mitochondrial membrane potential has previously been implicated as a biomarker or mediator of age-related cellular pathology⁴²⁻⁴⁶. By tracking the mitochondrial membrane potential in individual cells across the aging process, Fehrmann *et al.* found that the instantaneous risk of losing mitochondrial membrane potential did not increase with age. Moreover, they reported that the cells who eventually lost mitochondrial membrane potential were not significantly shorter-lived than those who maintained high mitochondrial potential for their entire lifespans¹¹. These types of observations are wholly dependent on the ability to follow single cells and observe their physiological states through the entire aging process.

4. Considerations

Because lingering progeny cells will divide and proliferate exponentially, microfluidic lifespan devices must effectively clear the vast majority of daughter cells to keep the device from becoming clogged with cells. Thus, a very delicate balance must be struck between retention of mother cells and removal of unwanted progeny. The key innovation of most of these devices is a geometry that exploits the size difference between mother and daughter cells to immobilize aging mothers and wash away new daughter cells. And as such, all designs are very sensitive to the size of the mother. All devices mentioned in this review have focused on haploid cells, and although the size difference and altered budding pattern of diploids doesn't ensure failure, so far none of these devices have been tested on diploid cells. While this may not be a severe limitation to the utility of such devices, it is clear that ploidy can impact replicative lifespan in yeast (for example BY4743 diploids are longer-lived than the corresponding haploid strains)⁴⁷, and aging studies in nearly every other model system are performed in diploid organisms.

Moreover, even within haploids, this balance is complicated by several factors. Mother and daughter cells are nearly identical in size during the first division or two. The device needs to be able to trap these small mother cells effectively at the beginning of the experiment. However, over the course of their lifespans, old mother cells can grow to be multiple times the size of young mother cells. The trapping geometry needs to accommodate these much larger cells as well, as excessive pressure on cells can induce stress responses and may reduce lifespan^{9,14}. Daughters of old mother cells are also larger⁴⁸, and it is unknown whether pressure on growing buds (constrained between two columns in some designs) can affect the aging process. Additionally, a large daughter bud can dislodge its mother from a trap, and multiple buds sometimes remain attached to the mother cell in a clustered arrangement, further complicating the designs. Judgment of the success of many of the published devices in overcoming these obstacles is difficult due to the lack of raw data available (i.e. time-lapse videos showing the whole device or an entire field of view). Indeed, the devices mentioned in this review have widely varying trends for mother cell retention through time (see **Table 1**). Lastly, because the devices feature differences in general layout as well as in trap shape, the same trap design may perform differently depending on the overall device layout.

An experiment where most of the cells are lost before senescence is not necessarily a failure. Statistical right-censoring to utilize partial lifespan data (i.e. a mother cell that is retained for several divisions but washed out before senescence) is routinely used in survival analysis⁴⁹. However, the reasoning that undergirds this method assumes that cells disappearing before senescence are not systematically different than those trapped for their entire lifespans. The fact that these devices exploit cellular size or budding characteristics to trap mother cells is a strong indicator that this requirement may not be upheld. Moreover, as cells continue to grow in volume over their entire lifespans, published work has suggested that cell size may be a biomarker of remaining lifespan^{13,50}. Thus genetic or environmental interventions that affect lifespan through pathways that also affect cellular budding patterns or geometry may not be suitable for study in some devices. This may be a particularly important limitation, given that many genetic and environmental interventions that extend lifespan also affect mother cell size^{50,51}.

Thus far, studies have not attempted to demonstrate robustness of lifespan results to microfluidic-specific experimental conditions such as fluid speed, culture growth phase, or pre-loading treatments

such as sonication or centrifugation. Some of the published devices have reported lifespans both for wild-type cells, and a small number of short- and long-lived strains that are comparable to those measured by microdissection^{8-10,13,14}. However, a major limitation of the current literature in this area is that no studies have assessed the ability of any microfluidic device to recapitulate the reported longevity effects of a large number of longevity-altering interventions. In one case, Huberts *et al.* reported that the longevity-promoting effect of caloric restriction is not seen in their microfluidic design. Their observations were accompanied by an interesting meta-analysis showing that studies showing the protective effects of dietary restriction in yeast generally reported control populations that were shorter-lived than the norm⁵². Later, Jo *et al.* found that dietary restriction does increase longevity in cells aged in their device, noting that the conclusions of Huberts *et al.* were drawn from lifespan experiments in which ~85% of initial population was censored¹³.

Going forward, it will be important to establish which microfluidic device designs, and under what experimental conditions, most closely recapitulate the results obtained using the “gold standard” microdissection assay across a spectrum of genetic and environmental conditions. This is particularly true in those cases where the yeast replicative aging model has been found to recapitulate the effects of analogous interventions in multicellular eukaryotes, such as *C. elegans*^{53,54}. Key pathways that should be explored in detail include the ability of yeast cells to show lifespan extension from dietary restriction (both glucose and other nutrients) and the response to conserved genetic modifiers of aging such as sirtuins and the target of rapamycin (TOR).

5. Dissemination

In addition to concerns about device performance, adoption of microfluidic technologies can be difficult for labs without previous microfluidics expertise. While the scientists who develop and use these devices are generally happy to share blueprints of the photomask (which can then be purchased from a number of sources), in order to manufacture the device molds using the photomasks, a nanofabrication facility is typically required. In addition, there are often significant knowledge and financial barriers for labs to set up and troubleshoot the microfluidic system even after obtaining a working device. For example, use of these devices has been largely limited to laboratories with access to expensive motorized microscopes. These microscopes allow researchers to parallelize experiments, capturing lifespans of multiple strains at once (through device design or the use of multiple devices at once). Most yeast labs do not have such a microscope available that can be dedicated solely to yeast lifespan experiments and, because experiments require continuous monitoring for upwards of 2 days, core facility rates for such a microscope can also be unaffordable.

In order to help overcome this barrier, we have set out to develop a lower cost microfluidic platform for basic analysis of replicative lifespan and cell morphological features during aging. We are currently optimizing a system that costs about \$2,500 to build from scratch. This includes the microscope, camera, syringe pump, and additional necessary accessories to perform microfluidic aging experiments. We anticipate publication and dissemination of this system within the next year, and we hope this will facilitate broader adoption of microfluidic technologies within the yeast geroscience community.

6. Conclusion and Future

The reviewed microfluidic devices offer alternatives to the traditional method of microdissection for collecting replicative lifespan data. Moreover, these devices are currently the only validated methods for collecting *in situ*, single-cell data on aging physiology across an entire lifespan. Adoption of these methods can be challenging but has begun to spread beyond the labs that developed each design, and they are poised to become an essential tool for the study of replicative aging in yeast. The impact of different device designs and experimental conditions on robustness and reproducibility is currently unknown, however, and this should be a major focus of laboratories designing and utilizing microfluidic systems for yeast aging in the future.

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FIGURE LEGENDS

Figure 1: Microfluidic device designs. A) This device relies both on a shortened confining ceiling, and chemical bonds. The surface of the glass is coated in streptavidin, and all the mother cells are labeled with biotin. This not only confines the cells vertically, but bonds them chemically to the surface. B) Two groups independently developed devices that rely solely on confinement under small columns or micropads. These small regions physically restrain the larger mothers, but allow the newborn daughters to wash away. C) CLiC device. This constrains the cells in both the vertical dimension, and places the mother cell in a narrow, elongated cavity in the wall. A small confluent monolayer of cells grows around the cavity, but once the region is full, cells are pushed out and washed away by the bypass channels. D) ALCATRAS device design. A small set of PDMS pillars in a V-shape traps the larger mother cells, and allow daughters to bud through the opening and be washed away. E) HYAA device design. A pair of L-shaped PDMS pillars traps mother cells and allows daughters to be removed by flow. F) Yeast Replicator device design. A small PDMS jail composed of three square pillars traps small newborn daughters and holds them. New daughters bud outside the trap and are washed away.

Table 1: Key differences between microfluidic devices. *Actual number of trapped mother cells/field of view is highly dependent on loading protocol including density of seeding culture, injection pressure, and length of seeding period. Often, optimal filling of traps must be balanced against having too many cells per trap or a greater chance of cells caught in non-trap areas leading to overgrowth of device.

Publications Grouped by Device	Trapping Method	Strains per Device	Mother Retention	Ensure Virgins	Single Trap Footprint*	Mutant Strains Tested for Lifespan	Raw Data Provided
Xie et al. 2012 Rafelski et al. 2012	Height restriction + Chemical	8	Not Reported	No	N/A, "3-5 cells per field of view"	<i>fob1Δ</i> <i>ypt11Δ</i>	Videos of entire field of view
Lee et al. 2012 Huberts et al. 2014	Pensile Column/ Micropad	1	~15-30%	No	Single pad: 450 μm^2 With spacing: ~850 μm^2 (up to 2 cells/trap)	<i>fob1Δ</i> <i>sir2Δ</i>	Videos of single traps
Zhang et al. 2012 Xie et al. 2015	Pensile Column/ Micropad	4	Not Reported	No	Single column: 1600 μm^2 Spacing not reported (up to 2 cells/trap)	<i>est2-D530A mrc1^{AQ}tlc1Δ</i> <i>fob1Δ rad9Δtlc1Δ</i> <i>mrc1^{AQ} rad52Δsml1Δ</i> <i>sir2Δ sml1Δtlc1Δ</i> <i>sml1Δ tel1Δtlc1Δ</i> <i>tel1Δ mrc1^{AQ}Δtel1Δtlc1Δ</i> <i>tlc1Δ rad52Δsml1Δtlc1Δ</i> <i>mec1Δsml1Δ</i>	Videos of single traps
Fehrmann et al. 2013 Meitinger et al. 2014	Elongated Cavities	1	~65%	Yes	Single trapping area: ~3840 μm^2 (up to 2 cells/trapping area)	<i>nba1Δ</i> <i>rga1Δ</i> <i>nba1Δrga1Δ</i>	Videos of single traps
Crane et al. 2014	2-Column Traps	1	~40-60%	No	Single trap: ~100 μm^2 , With spacing: ~750 μm^2	None	Video of entire chip
Jo et al. 2015	2-Column Traps	16	>90%	No	Single trap: ~200 μm^2 With spacing; ~700 μm^2	<i>bre1Δ rpn4Δ</i> <i>chl1Δ sas2Δ</i> <i>fob1Δ sip2Δ</i> <i>hsp104Δ tma19Δ</i> <i>idh1Δ tor1Δ</i> <i>rpl22aΔ ubr2Δ</i>	Videos of single traps
Liu et al. 2015	3-Column Jails	10	Not Reported	Yes	Single trap: ~175 μm^2 With spacing: 1788 μm^2	<i>fob1Δ sir2Δ</i> <i>sgf73Δ gpa2Δ</i> <i>tor1Δ rpl31aΔ</i>	Photos of single cells of unknown age

Table 2: Reported lifespans of wild-type haploid cells in each microfluidic device. *Approximate values are estimated from survival curves. †Includes all details specified in the publication

Reporting Publication	Trapping Principle	Running Media	Growth Conditions Prior to Loading [†]	Strain	Lifespan*
Xie et al. 2012	Mechanical + Chemical	YPD	grown in YPD	BY4741	median $\approx$ 17
Lee et al. 2012	Pensile Column/ Micropad	YPD or Synthetic Defined with 2% glucose	grown overnight to stationary phase, diluted to OD ₆₀₀ = 0.3, incubated 3h at 30 °C, sonicated mildly	“BY”	median = 25
Zhang et al. 2012	Pensile Column/ Micropad	Not Reported	grown in YPED at 30 °C overnight to OD ₆₀₀ = 1.0, diluted 1: 50, incubated 6 h at 30 °C	BY4741 BY4742	BY4741: mean = 23.6 BY4742: mean = 26.0
Fehrmann et al. 2013	Elongated Cavities	Synthetic Complete with 2% glucose (SC)	“... grown overnight, transferred to fresh medium the next morning, and allowed to grow to log phase until transfer to the microfluidic device in the afternoon.”	BY4741	Including right-censored cells: median = 32.4 Including only cells seen to die: median = 25.3
Crane et al. 2014	2-Column Traps	XY with 2% glucose	grown overnight in XY with 2% glucose “and then diluted and allowed to grow to log phase...”	S288C	mean = 22.4
Jo et al. 2015	2-Column Traps	YPD, Synthetic Complete with 2% glucose (SC)	“... cultured in YPD at 30 °C overnight before diluting 10–50-fold and loading...”	BY4741	YPD: mean = 26.76 SC: mean = 24.16
Liu et al. 2015	3-Column Jails	YPD, CSM with 2% glucose	grown overnight in CSM with 2% glucose to final density of 0.25	“BY”	YPD: mean = 22.2 CSM: mean = 24.2

Table 3: Reported lifespans for various haploid strains in the S288C/BY background measured in microfluidic devices. Microfluidic lifespan values to the tenth precision indicate mean. Whole number indicates median. Tilde indicates median estimated from survival curve. Microdissection lifespan ranges are reported as mean and were obtained from published studies in the BY4742 strain background ^{6,47,54-63}.

Strain	Microfluidic Lifespan	Microdissection Lifespan
<i>bre1Δ</i>	17.2 (Jo et al. 2015)	17.6
<i>chl1Δ</i>	19.3 (Jo et al. 2015)	16.0
<i>est2-D530A</i>	7.6 (Zhang et al. 2015)	
<i>fob1Δ</i>	~29 (Xie et al. 2012) 33.1 (Zhang et al. 2012) 33.9 (Jo et al. 2015) 31.0 (Liu et al. 2015)	27.4-37.8
<i>gpa2Δ</i>	29.9 (Liu et al. 2015)	32.0-36.7
<i>hsp104Δ</i>	26.0 (Jo et al. 2015)	32.4
<i>idh1Δ</i>	27.9 (Jo et al. 2015)	30.5-33.5
<i>mrc1^{AQ}</i>	~25 (Zhang et al. 2015)	
<i>nba1Δ</i>	14 (Meitinger et al. 2014)	
<i>rga1Δ</i>	12 (Meitinger et al. 2014)	
<i>rpl22aΔ</i>	30.0 (Jo et al. 2015)	35.8-39.8
<i>rpl31aΔ</i>	30.6 (Liu et al. 2015)	34.6-35.5
<i>rpn4Δ</i>	18.1 (Jo et al. 2015)	13.3-17.3
<i>sas2Δ</i>	31.8 (Jo et al. 2015)	28.4-32.4
<i>sgf73Δ</i>	29.6 (Liu et al. 2015)	42.2
<i>sip2Δ</i>	28.6 (Jo et al. 2015)	31.1
<i>sir2Δ</i>	15.4 (Zhang et al. 2012) 13.3 (Jo et al. 2015) 12.7 (Liu et al. 2015)	12.4-14.0
<i>sml1Δ</i>	~27 (Zhang et al. 2015)	
<i>tel1Δ</i>	~27 (Zhang et al. 2015)	
<i>tlc1Δ</i>	12.6 (Zhang et al. 2015)	
<i>tma19Δ</i>	27.3 (Jo et al. 2015)	33.4
<i>tor1Δ</i>	29.0 (Jo et al. 2015) 30.7 (Liu et al. 2015)	28.1-33.2
<i>ubr2Δ</i>	31.2 (Jo et al. 2015)	35.9-42.5
<i>ypt11Δ</i>	Bimodal, overall mean unspecified (Rafelski et al. 2012)	
<i>mec1Δsml1Δ</i>	~20 (Zhang et al. 2015)	
<i>mrc1^{AQ}tlc1Δ</i>	8.8 (Zhang et al. 2015)	
<i>nba1Δrga1Δ</i>	6 (Meitinger et al. 2014)	
<i>rad9Δtlc1Δ</i>	16.5 (Zhang et al. 2015)	
<i>rad52Δsml1Δ</i>	~12 (Zhang et al. 2015)	
<i>sml1Δtlc1Δ</i>	~22 (Zhang et al. 2015)	
<i>tel1Δtlc1Δ</i>	9.8 (Zhang et al. 2015)	
<i>mrc1^{AQ}Δtel1Δtlc1Δ</i>	11.5 (Zhang et al. 2015)	
<i>rad52Δsml1Δtlc1Δ</i>	~7 (Zhang et al. 2015)	