Fig. 2 Effect of mutant and wildtype DAF-16a::GFP proteins on lifespan. $P$ values were calculated for individual experiments, each consisting of control and experimental animals examined at the same time. We show the cumulative statistics because we observed that the strains behaved similarly in different experiments (we observed no statistically-significant differences). n , total number of animals observed (the number of independent experiments performed is given in parentheses); $m$, mean lifespan $\pm$ SEM; $P$, p values. $\boldsymbol{a}, \mathrm{N} 2, \mathrm{n}=348(5), m=19.4 \pm 0.3$; daf-2(e1370), $\mathrm{n}=140(2), m=$ $45.8 \pm 1.5, P<0.0001$ vs. N2; daf-16(mu86); daf-2(e1370), $\mathrm{n}=140(2), m=14.8 \pm 0.4, \mathrm{P}$ < 0.0001 vs daf-2(e1370) or N2; daf-16(mu86); daf-2(e1370); muIs61, $\mathrm{n}=37$ (1), $m=$ $44.1 \pm 3.6, P<0.0001$ vs. daf-16(mu86); daf-2(e1370) or N2. $\boldsymbol{b}$, daf-16 (mu86); daf2(e1370); muEx108, $\mathrm{n}=49$ (1), $m=40.1 \pm 3.3, P=0.075$ vs. $d a f-2(e 1370), P<0.0001$ vs. daf-16 (mu86); daf-2(e1370) or N2; daf-16 (mu86); daf-2(e1370); muEx110, $\mathrm{n}=70$ (1), $m=38.5 \pm 2.4, P=0.018$ vs. $d a f-2(e 1370) ; P<0.0001$ vs. $d a f-16$ (mu86); daf2(e1370) or N2. c, N2 carrying muEx167, $\mathrm{n}=49(1), m=20.8 \pm 0.9, P=0.047$ vs. N2; N 2 carrying $m u E x 107, \mathrm{n}=50(1), m=21.2 \pm 0.8, P=0.020$ vs. N2; N2 carrying $p R F 4$ alone, $\mathrm{n}=190(3), m=18.4 \pm 0.4, P=0.286$ vs. N2. $\boldsymbol{d}$, N 2 carrying $m u E x 53, \mathrm{n}=60(1)$, $m=18.7 \pm 0.9, P=0.83$ vs. N2; N2 carrying muEx58, $\mathrm{n}=61(1), m=21.2 \pm 0.9, P=$ 0.0006 vs. $\mathrm{N} 2 ; \mathrm{N} 2$ carrying $m u E x 59, \mathrm{n}=67(1), m=22.1 \pm 0.9, P<0.0001$ vs. $\mathrm{N} 2 ; \mathrm{n}=$ 188 (3), $m=17.0 \pm 0.5, P=0.0035$ vs. N2. e, daf-16(mu86) $\# 1, \mathrm{n}=140(2), m=15.1 \pm$ $0.3, P<0.0001$ vs. N2; daf-16(mu86) $\# 2, \mathrm{n}=140(2), m=14.5 \pm 0.3, P<0.0001$ vs. N2, $P=0.36$ vs. $d a f-16(m u 86) \# 1 ; d a f-16(m u 86) ; d a f-2(e 1370), \mathrm{n}=140(2), m=14.8 \pm 0.4, P$ $<0.0001$ vs. N2, $P=0.61$ vs $d a f-16(m u 86) \# 1, P=0.54$ vs. $d a f-16(m u 86) \# 2 . f . d a f-$
$16(m u 86), \mathrm{n}=140(2), m=15.1 \pm 0.3, P<0.0001$ vs. N 2 ; daf-16(mu86); $m u E x 116, \mathrm{n}=$ 50 (1), $m=16.6 \pm 0.8, P=0.038$ vs. daf-16(mu86), $P=0.0002$ vs. N2; daf-16(mu86); $m u E x 160, \mathrm{n}=82(1), m=20.8 \pm 0.8, P<0.0001$ vs. $d a f-16(m u 86), P=0.051$ vs. N 2 ; daf-16(mu86); daf-2(e1370); muEx158, $\mathrm{n}=70$ (1), $m=31.7 \pm 1.9, P<0.0001$ vs. N 2 ; daf-16(mu86); daf-2(e1370); muEx159, $\mathrm{n}=65$ (1), $m=34.5 \pm 2.3, P<0.0001$ vs. N2. $\boldsymbol{g}$, daf-16(mu86); $m u E x 151, \mathrm{n}=90(1), m=17.9 \pm 0.9, P=0.0014$ vs. $d a f-16(m u 86), P=$ 0.32 vs. N2; daf-16(mu86); $m u E x 152, \mathrm{n}=100(1), m=21.2 \pm 1.0, P<0.0001$ vs. daf16(mu86), $P=0.026$ vs. N2; daf-16(mu86); muEx154, $\mathrm{n}=60$ (1), $m=22.7 \pm 1.3, P<$ 0.0001 vs. $d a f-16(m u 86) ; P=0.0005$ vs. N2; daf-16(mu86); daf-2(e1370); muEx151, n $=60(1), m=35.4 \pm 2.8, P<0.0001$ vs. N2; daf-16(mu86); daf-2(e1370); muEx154, $m=$ $34.9 \pm 2.8, P<0.0001$ vs.N2.

