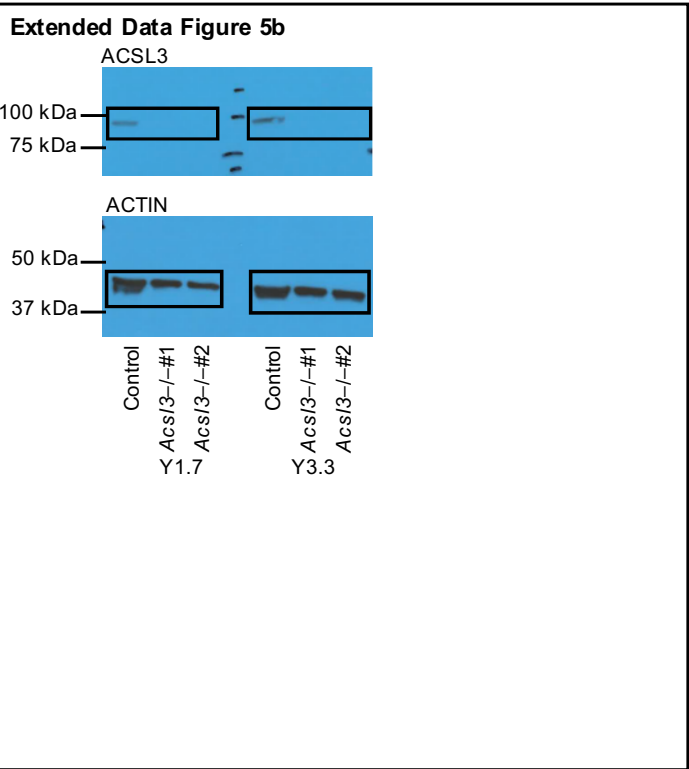
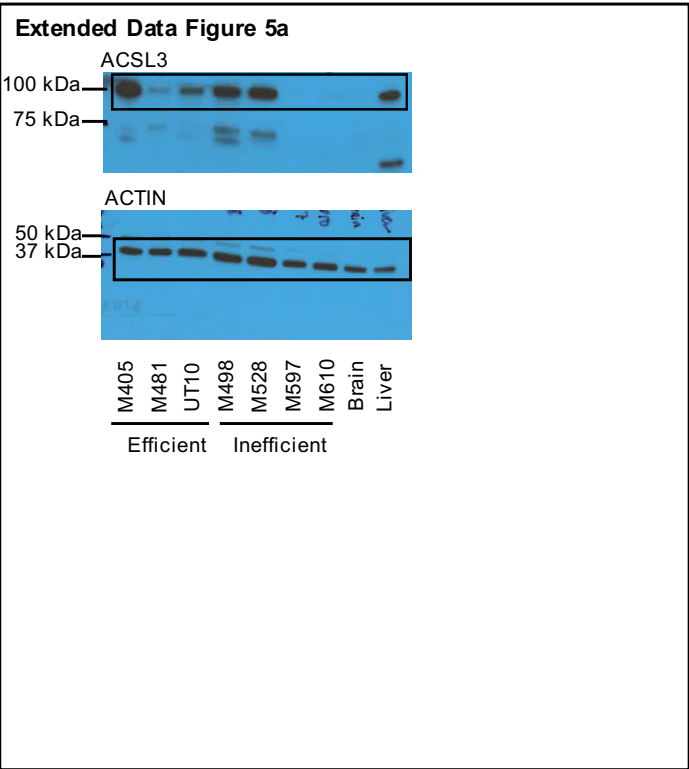
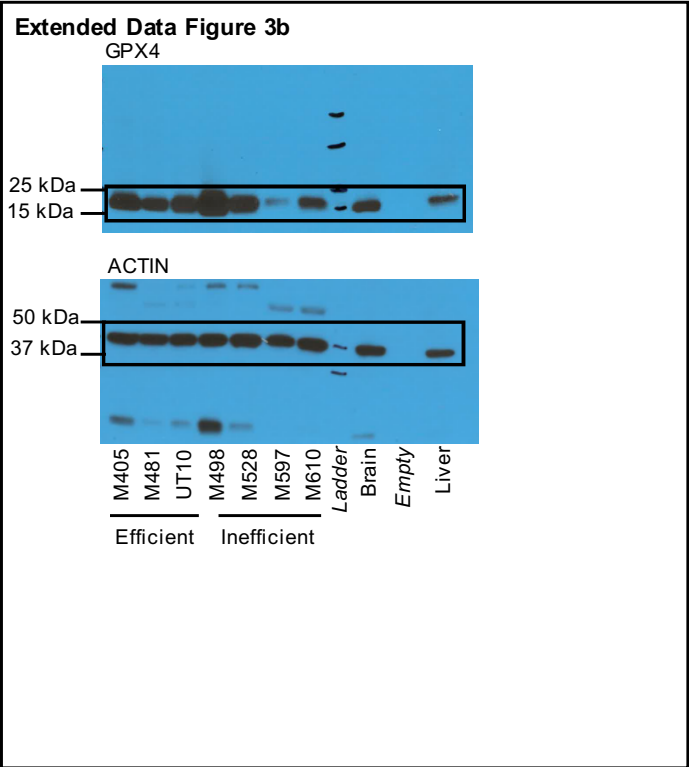
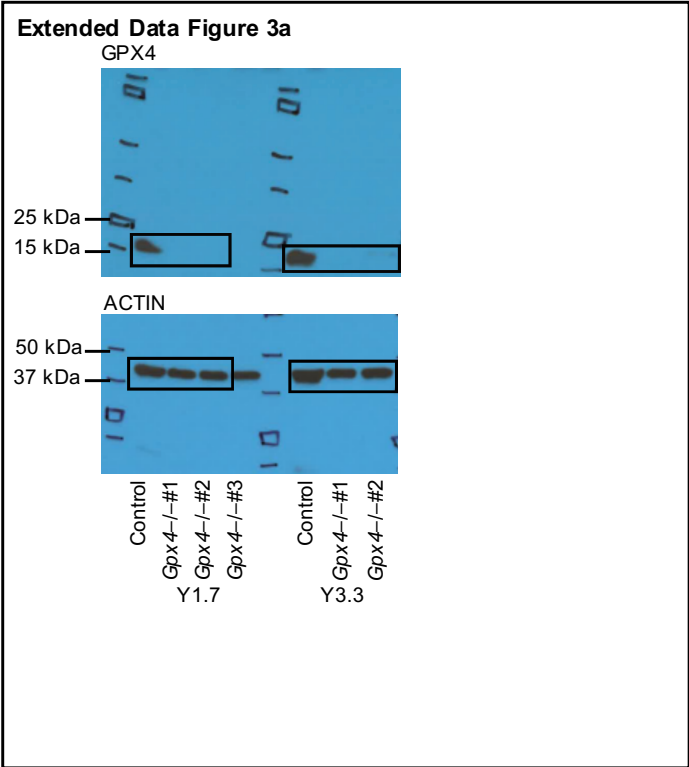
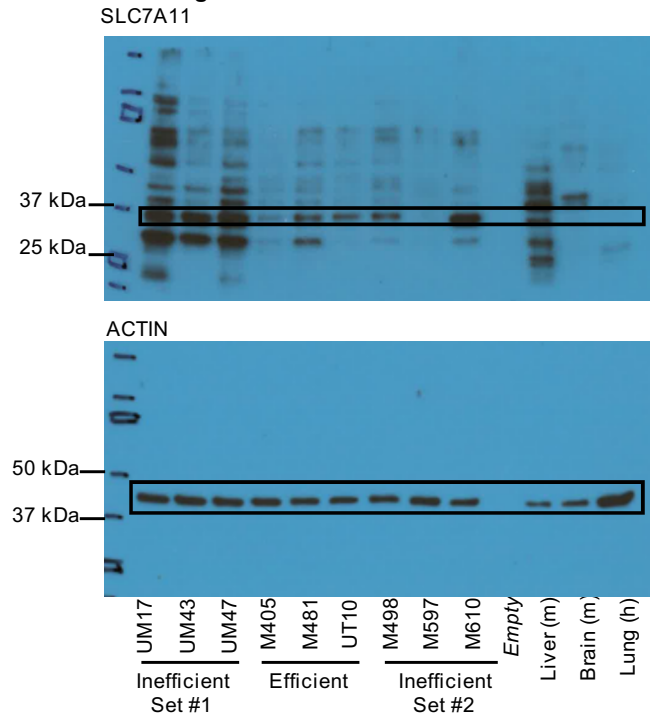

Supplementary information

Lymph protects metastasizing melanoma cells from ferroptosis

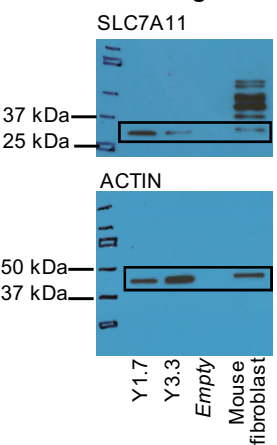
In the format provided by the
authors and unedited



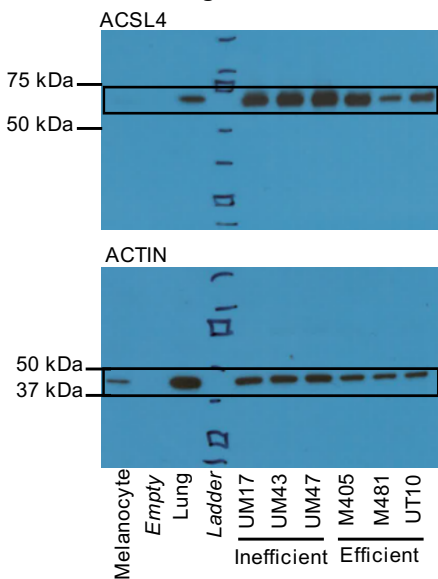
Extended Data Figure 6a



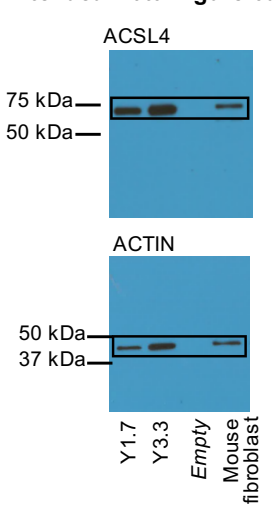
Extended Data Figure 6b



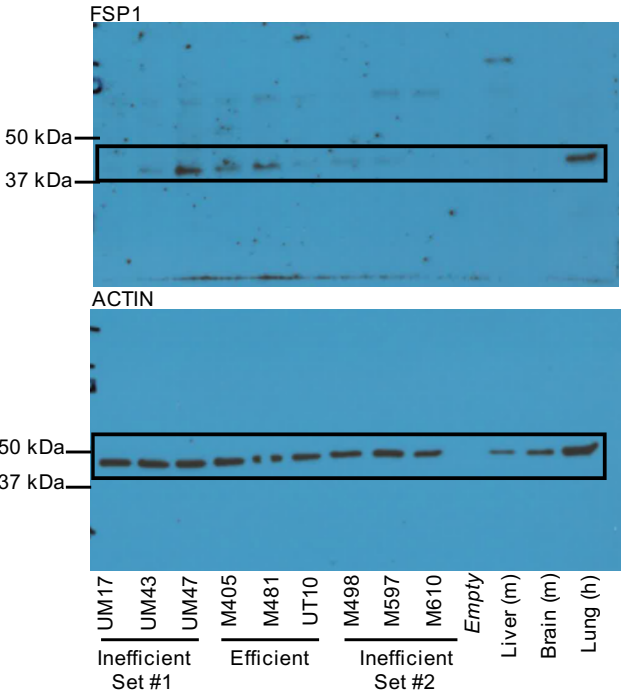
Extended Data Figure 6c



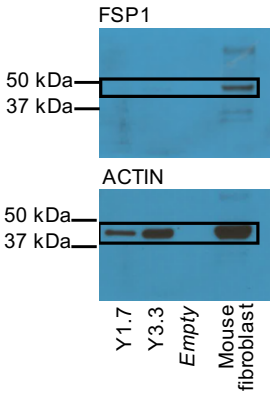
Extended Data Figure 6d



Extended Data Figure 6e



Extended Data Figure 6f



Supplementary Data.

STATISTICS AND REPRODUCIBILITY

Table 1.

These data reflect n=520 mice total (exact numbers of mice per melanoma per cell dose for each injection are in the source data file) from three biologically independent experiments per melanoma

Figure 1.

a, a total of n=4 replicates (each replicate was pooled from 6-10 mice) per melanoma from two biologically independent experiments per melanoma

b, a total of n=5 replicates (each replicate was pooled from 6-10 mice) per melanoma from two biologically independent experiments per melanoma

c, a total of n=9-10 (M405), n=3-6 (M481), or n=5 (UT10) replicates (each replicate was pooled from 6-10 mice) per melanoma from three (M405) or two (M481, UT10) biologically independent experiments per melanoma

d, a total of n=5 replicates (each replicate was pooled from 6-10 mice) per melanoma from two biologically independent experiments per melanoma

e, a total of n=3 (M405, M481) or n=5-6 (UT10) replicates (each replicate was pooled from 6-10 mice) per melanoma from two biologically independent experiments per melanoma

f, a total of n=3 replicates (each replicate was pooled from 6-10 mice) per melanoma from three biologically independent experiments per melanoma

g, a total of n=3 (NSG) or n=5 (C57BL) replicates (each replicate was pooled from 6-10 mice) from three (NSG) or two (C57BL) biologically independent experiments per mouse strain

h, a total of n=3 (NSG) or n=5 (C57BL) replicates (each replicate was pooled from 6-10 mice) from three (NSG) or two (C57BL) biologically independent experiments per mouse strain

i, a total of n=3 (NSG) or n=5 (C57BL) replicates (each replicate was pooled from 6-10 mice) from three (NSG) or two (C57BL) biologically independent experiments per mouse strain

j, a total of n=4 (NSG) or n=5 (C57BL) replicates (each replicate was pooled from 6-10 mice) from two biologically independent experiments per mouse strain

Figure 2.

a, a total of n=4 (M405), n=8 (M481), or n=3 (UT10) replicates (each replicate was pooled from 4-5 mice) per melanoma from two biologically independent experiments per melanoma

b, a total of n=5 (Y1.7) or n=6 (Y3.3) replicates (each replicate was pooled from 4-5 mice) per melanoma from three biologically independent experiments per melanoma

c, a total of n=20-24 cultures per treatment per melanoma from three biologically independent experiments per melanoma

d, a total of n=4 cultures per treatment per melanoma from one biologically independent experiment per melanoma; results are representative of two biologically independent experiments per melanoma

e, a total of n=5 replicates (each replicate was pooled from 4-5 mice) per mouse strain from one experiment per mouse strain; results are representative of two biologically independent experiments per mouse strain

f, a total of n=5 (M405, UT10), n=15 (M481, Control), n=20 (M481, Liprox) mice per treatment from one (M405, UT10) or three (M481) biologically independent experiments per melanoma

g, a total of n=17 (Y1.7, Control), n=14 (Y1.7, Liprox), n=25-26 (Y3.3) mice per treatment per melanoma from two (Y1.7) or three (Y3.3) biologically independent experiments per melanoma

h, a total of n=10 mice per treatment per melanoma from two biologically independent experiments per melanoma

i, a total of n=8-10 mice per treatment per melanoma from two biologically independent experiments per melanoma

j, a total of n=8-9 mice per treatment per melanoma from two biologically independent experiments per melanoma

k, a total of n=6-7 (Y1.7) or n=9-10 (Y3.3) mice per treatment per melanoma from two biologically independent experiments per melanoma

l, a total of n=10 mice per parental or knockout melanoma line from two biologically independent experiments per melanoma

m, a total of n=10 mice per parental or knockout melanoma line from two biologically independent experiments per melanoma

n, a total of n=10 mice per parental or knockout melanoma line from two biologically independent experiments per melanoma

Figure 3.

a, a total of n=6 replicates (each replicate was pooled from 4-5 mice) from three biologically independent experiments

b, a total of n=6 replicates (each replicate was pooled from 4-5 mice) from three biologically independent experiments

c, a total of n=24 (M405) or n=15 (M481, UT10) replicate cultures per treatment per melanoma from three (M405) or two (M481, UT10) biologically independent experiments per melanoma

d, a total of n=4 replicate cultures per treatment per melanoma from one experiment per melanoma

e, a total of n=10-20 (M405), n=15 (M481), or n=4-5 (UT10) mice per treatment per melanoma from four (M405), three (M481), or one (UT10) biologically independent experiments per melanoma

f, a total of n=10 mice per treatment per melanoma from two biologically independent experiments per melanoma

g, a total of n=5 replicates (replicates were pooled from 4-5 mice) per mouse strain from one experiment per mouse strain

h, a total of n=5 replicates (replicates were pooled from 4-5 mice) per mouse strain from one experiment per mouse strain

i, a total of n=28 (*Acs/3-/-*#1) or n=24 (*Acs/3-/-*#2) replicate cultures per treatment per cell line from three biologically independent experiments per cell line

j, a total of n=28 (*Acs/3-/-*#1) or n=28 (*Acs/3-/-*#2) replicate cultures per treatment per cell line from three biologically independent experiments per cell line

k, a total of n=10 mice per melanoma per injection type from two biologically independent experiments per injection type

l, a total of n=3 replicate cultures per treatment per melanoma from one experiment per melanoma; results are representative of two biologically independent experiments per melanoma

m, a total of n=3 replicate cultures per treatment per melanoma from one experiment per melanoma; results are representative of two biologically independent experiments per melanoma

Extended Data Table 1.

a, The number of total mice analyzed in each cohort are indicated in parentheses and the data reflect results from 1 to 5 biologically independent experiments per melanoma.

b, The number of mice analyzed in each cohort are indicated in parentheses and the data reflect results from two biologically independent experiments per melanoma.

Extended Data Figure 1.

e, This image is representative of over 25 independent experiments of metastatic tumor burden analysis by ex vivo bioluminescence imaging of organs from an uninjected NSG mouse.

f, This image is representative of over 25 independent experiments of metastatic tumor burden analysis by ex vivo bioluminescence imaging of organs from an NSG mouse xenografted with a patient-derived melanoma (M481)

g, This image is representative of over 25 independent experiments of metastatic tumor burden analysis by ex vivo bioluminescence imaging of organs from a C57BL mouse injected

subcutaneously with a mouse melanoma (Y3.3)

i, a total of n=4 (M597) or n=5 (UM43, UM47) replicates (each replicate was pooled from 6-10 mice) per melanoma from two biologically independent experiments

Extended Data Figure 2.

a, a total of n=3 replicate mice (tumor and blood from the same mouse) from one experiment

b, a total of n=3 replicate cultures per treatment per melanoma from one experiment

c, a total of n=5 replicate cultures per treatment per melanoma from one experiment

d, a total of n=5 mice per treatment; data are from one experiment that is representative of two biologically independent experiments

e, a total of n=5 mice per treatment; data are from one experiment that is representative of two biologically independent experiments

f, a total of n=5 mice per treatment; data are from one experiment that is representative of two biologically independent experiments

g, a total of n=7 (control) or n=10 (Liprox) mice per treatment; data are from one experiment that is representative of two biologically independent experiments

h, a total of n=7 (control) or n=9 (Liprox) mice per treatment; data are from one experiment that is representative of two biologically independent experiments

i, a total of n=5 mice per treatment per melanoma from one experiment

j, a total of n=20 (Y1.7) or n=10 (Y3.3) mice per treatment per melanoma from two biologically independent experiments

Extended Data Figure 3.

a, results are representative of two biologically independent experiments

b, results are representative of two biologically independent experiments

c, a total of n=6 replicate cultures per melanoma per experiment from two biologically independent experiments; results are representative of four biologically independent experiments

d, a total of n=6 replicate cultures per melanoma per experiment from two biologically independent experiments; results are representative of four biologically independent experiments

e, a total of n=6 subcutaneous tumors per cell line from two biologically independent experiments

f, a total of n=8-10 mice per cell line from two biologically independent experiments

g, a total of n=10 mice per cell line from two biologically independent experiments

h, a total of n=8-10 mice per cell line from two biologically independent experiments

i, a total of n=10 mice per cell line from two biologically independent experiments

Extended Data Figure 4.

a, a total of n=5 replicates (replicates were pooled from 4-5 mice) per mouse strain from two biologically independent experiments

b, a total of n=5 replicates (replicates were pooled from 4-5 mice) per mouse strain from two biologically independent experiments

c, a total of n=2 replicates (replicates were pooled from 4-5 mice) per mouse strain from two biologically independent experiments

d, a total of n=2 replicates (replicates were pooled from 4-5 mice) per mouse strain from two biologically independent experiments

Extended Data Figure 5.

a, results are representative of two biologically independent experiments

b, results are representative of two biologically independent experiments

c, a total of n=3-7 replicates per control or genetically modified melanoma from two biologically independent experiments

d, a total of n=3-6 replicates per control or genetically modified melanoma from two biologically independent experiments

e, a total of n=6 subcutaneous tumors per control or knockout melanoma line from two biologically independent experiments; data from parental control cells in this experiment are also shown in Extended Data Fig. 3e.

f, a total of n=4 replicate cultures per melanoma per experiment from one experiment; results are representative of two biologically independent experiments

g, a total of n=29-30 replicates per treatment per control or genetically modified melanoma line from three biologically independent experiments

h, a total of n=18-21 replicates per treatment per control or genetically modified melanoma line from two biologically independent experiments

i, a total of n=10 mice per control or knockout melanoma line from two biologically independent experiments

Extended Data Figure 6.

- a**, results are representative of two biologically independent experiments
- b**, results are representative of two biologically independent experiments
- c**, results are representative of two biologically independent experiments
- c**, results are representative of two biologically independent experiments
- e**, results are representative of two biologically independent experiments
- f**, results are representative of two biologically independent experiments
- g**, a total of n=3 replicates (replicates were pooled from 4-5 mice) per melanoma from one experiment
- h**, a total of n=2 subcutaneous tumors per melanoma except for UT10 (n=3 subcutaneous tumors) from one experiment
- i**, results are representative of two tumors per melanoma in two biologically independent experiments

In all instances, the indicated replicates are biologically independent samples from different mice or different cultures.

“Biologically independent experiments” are defined as experiments that were set up at different times in different mice or different cultures.