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#### SUPPLEMENTARY MATERIALS AND METHODS

#### Primers used for ChIP-qPCR

#### U6

GGCGCCAGTGCTCACTACTT GGGCCATGCTAATCTTCTCTG amplify - 87 to +61 from snRNA:U6:96Ac transcript start

### InR

P1 CATTCAGTGTTTCACGCGAT GGCAATTCACTGTGGCCTCT amplify -38844 to -38720 relative to translational start P3 GCCGCTGACAATTATTGTTGT GCCCGTGATTCGTGTGAGTGA amplify -9830 to - 9696 CDS GCGATGATCGCTGGAAGGTT GCGTTTGTCAATTCCGAGGAT amplify +2979 to +3105

#### Akt

GCAAGGGCACCTTTGGTAAG GTGGACTTGAGCACACGACTC amplify +885 to +1029

#### Cat-Indy

GCTTAGCCTACCGCTATTTG CTAGACTGAGAGAACTTAC amplify 3' past Cat and in the 3' end of Indy +17340 to +17219 relative to Indy start

#### TOR

ACGGATTCCTCGGACTTGGA TTTGGCCTCTGTACCGAAGT amplify from +4966 to + 5088

#### Su(Hw)

CACCCGCTCGTACCACTTGAA TACTCCGGCTTTTTAACCACCTGT amplify +1136 to +1304

#### Aldh-III

AAAATAATAATGGGTGTGGCTAAA GCGAAACGGGGGGAATAAT amplify +7477 to 7665

#### CG14131/Pi3K68D

CAGGCCGTTCTTGTTGTTGG ACGTGGCGGCGGTTGTT amplify from +93 to + 263. This (as well as the whole CG14131) is within the Pi3K68D (Phosphotidylinositol 3 kinase 68D; CG11621) gene.

#### CPTI

ATCTTGTATCGGACGGGTAATGA TGGGACAATGGGTGATAGGTG from +1954 to +2057

**4E-BP** (Thor) CGGCAATAACAACAAGAACCAG CGCCTGATTATTCTGCTTGGTC amplify -367 to -267

### SUPPLEMENTARY FIGURES

### CAPTIONS

**Supplementary Figure 1** Example of signal reproducibility in the three independent biological repeats of ChIP-chip on wild-type untreated females. The top panel shows the processed combined data with the peaks called indicated in colour, the lower three panels show raw (scaled log 2 Cy5/Cy3 ratios) data for the individual repeats.

**Supplementary Figure 2** Pearson correlation coefficients for all the ChIP-chip experiments. The raw scaled data for all ChIP-chip experiments were compared to each other and the Pearson correlation coefficients determined. The values of the Pearson correlation coefficients are indicated by the intensity of red (see scale). The experiments are coded in the legend. The mock-controls (IP with pre-immune serum on wild-type chromatin and anti-dFOXO IP on *dfoxo* null chromatin) are less similar to the experimental samples than the experimental samples are amongst themselves. Unstressed wild-type, stressed wild-type and  $daGAL4>UAS-dInR^{DN}$  samples are all highly similar.

**Supplemental Figure 3** Analysis of dFOXO distribution with respect to gene features on a genome-wide scale. Upstream/downstream refer to 1 kb from the 5'/3' UTR. Z-score represents the number of standard deviations each observation is from the mean of the distribution of  $10^3$  random peak sets, of identical size, length and chromosomal distribution. dFOXO bound regions are over-represented in genic regions, and tend to localise to the 3'-end of genes (p< $10^{-15}$ ), are not enriched in 5' UTR, and are underrepresented in 5' upstream region (p= $1.5 \times 10^{-4}$ ).



Supplementary Figure 1

# Supplementary Figure 2











### SUPPLEMENTARY TABLES

## Supplementary Table 1 De novo motif identification from peaks (MEME)

Motif consensus	р
GCTGCTGCTG	$1.3 \times 10^{-27}$
CTGCTGCTGC	$1.2 \times 10^{-14}$
CTCCTCCTCC	$1.3 \times 10^{-15}$
C[T/A]CCGCCGCC	$5.1 \times 10^{-13}$
CAGC[A/T]CTGC	$1.9 \times 10^{-8}$

**Supplementary Table 2** Overlap in dFOXO binding locations under standard conditions, compared to those under stress and dFOXO binding locations in daGAL4 UAS-InR<sup>DN</sup> flies compared to daGAL4 controls. For each comparison the overlap is given per set. The counts of peak overlaps suggest that there are unique peaks between the sets, however visual inspection of 'unique' treatment peaks in the control data shows in the great majority of cases that a peak of smaller magnitude exists at a level below the peak-calling threshold. For Pearson correlations amongst these experiments see Supplemental Figure 2.

Comparison	Peak set	Total peaks that directly overlap other peak set	Total peaks in set
Untreated vs	Untreated	1017	1423
Paraquat	Paraquat	957	1439
Untreated vs	Untreated	1098	1423
Starvation	Starvation	1078	1528

daGAL4	daGAL4	993	1455
VS			
daGAL4 UAS-	daGAL4 UAS-	1076	1643
$InR^{DN}$	$InR^{DN}$		