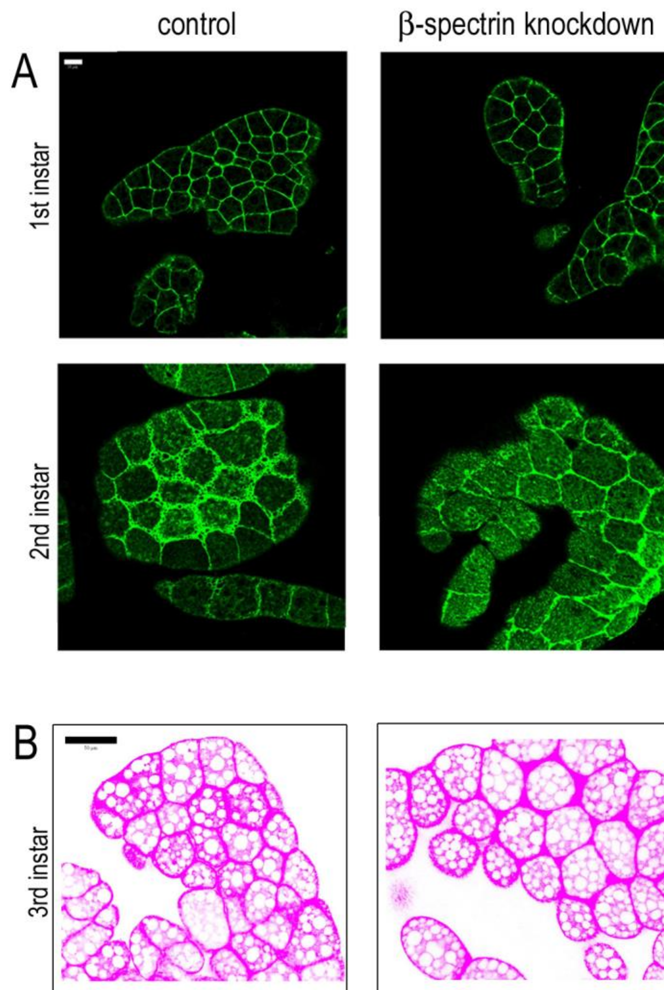

Figure S1 β spectrin knockdown did not affect the size and shape of fat body cells, or the accumulation of large lipid droplets. A. The pattern of CD8-GFP fluorescence was essentially the same in wild type and β spectrin knockdown fat body cells at 1st instar and at 2nd instar, when cortical lipid droplets normally begin to appear. B. Displaying an overexposed image of CD8-GFP fluorescence as a negative helped to highlight the pattern of large lipid droplets at 3rd instar and showed that their size and number were similar to wild type after β spectrin knockdown. Bar = 1 μm in A, 0.5 μm in B.

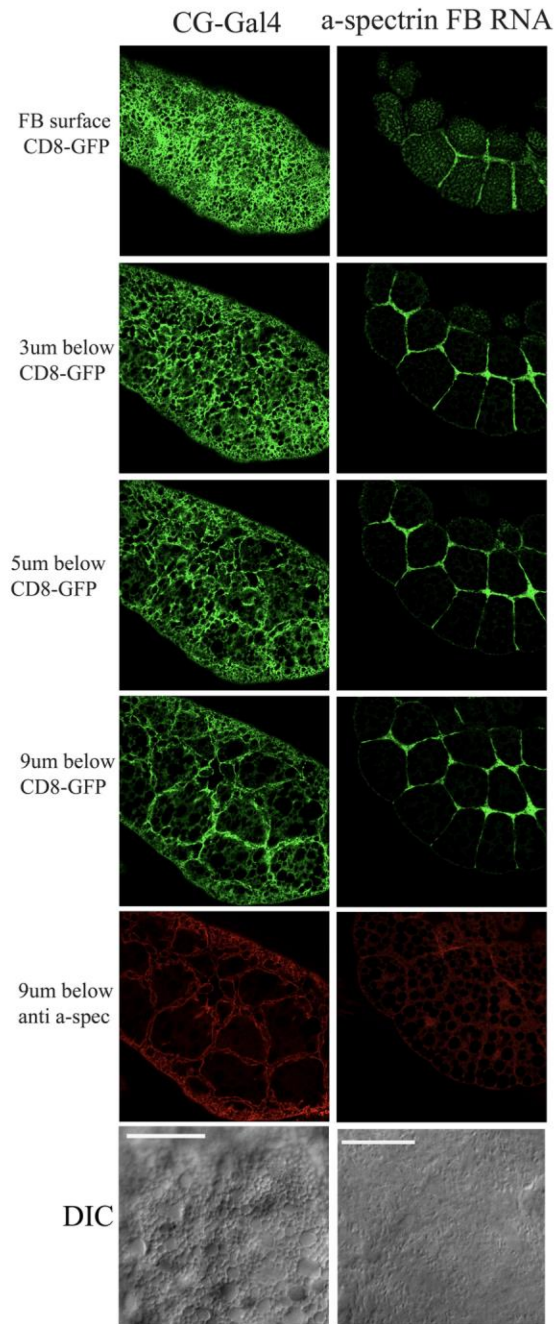

Figure S2 Knockdown of α spectrin produced the same effects on fat body morphology as β spectrin knockdown. The foamy appearance of the fat body surface detected via CD8-GFP was replaced by a speckled pattern on the ecto domain, as was observed with β spectrin RNAi. There was a concomitant loss of cortical lipid droplets as detected by DIC. Bar = 20 μm.

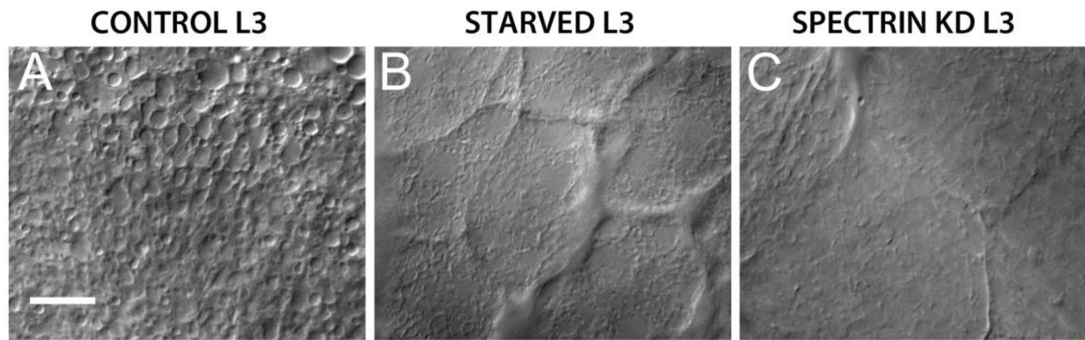

Figure S3 Cortical lipid droplets were lost from larval fat body during starvation. The population of small lipid droplets seen in the cortical region of fat body from wild type 3rd instar larvae (A) were nearly absent after starvation (B) as found with β spectrin knockdown with dsRNA (C). Bar = 20 μm.

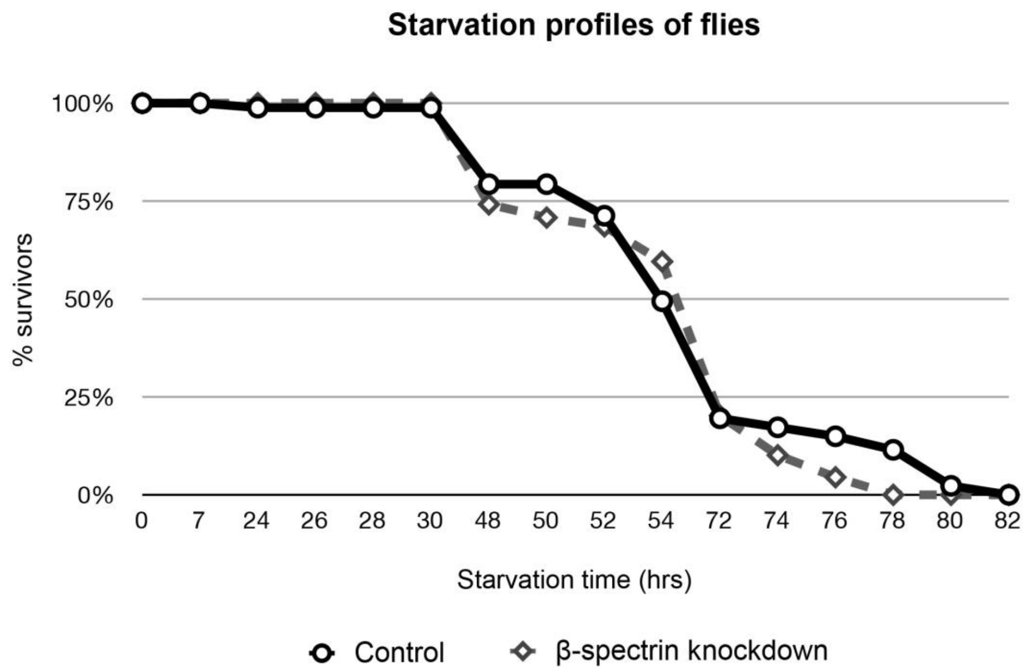

Figure S4 Starvation susceptibility profiles of control and β spectrin knockdown adults. Adult flies less than 24 hrs of age were transferred to agar vials containing water but no nutrients and kept at 25°C. 87 control (parental lines: UAS *βSpec*^{dsRNA} and *Cg*-Gal4) and 89 β spectrin knockdown adults (UAS *βSpec*^{dsRNA} X *Cg*-Gal4) were analyzed. Starvation susceptibility was tabulated as per cent survivors at the indicated times. It was previously shown that increased starvation susceptibility in *Lsd-2* mutants reflects a diminution of triacylglycerol stores compared to wild type (Gronke et al 2003). No such change was observed here with loss of spectrin function, suggesting that triacylglycerol stores were not altered. Thus, there appear to be effects of Lsd-2 beyond their requirement for cortical lipid droplet formation.

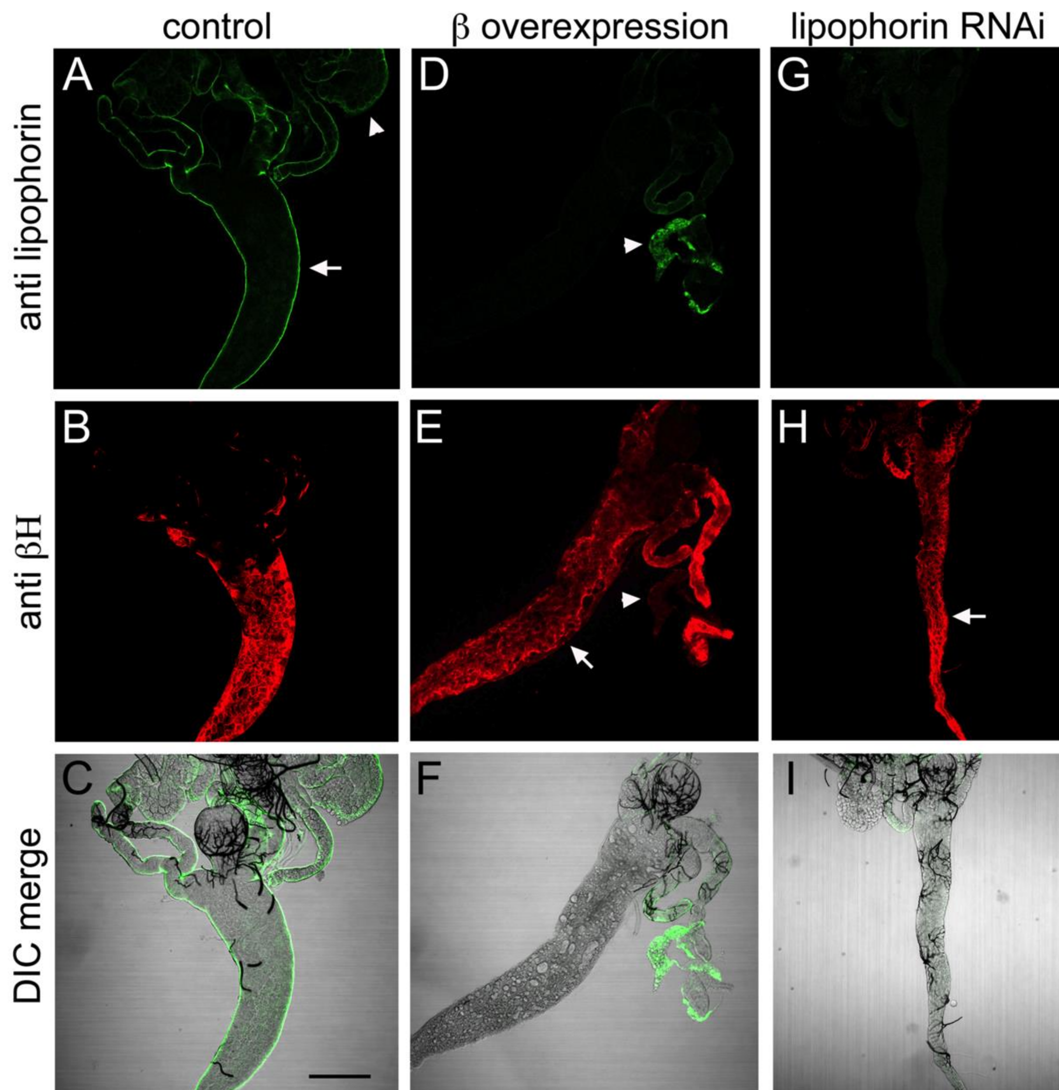

Figure S5 β spectrin overexpression in the larval fat body resulted in a loss of lipophorin accumulation at the midgut, comparable to that observed with lipophorin RNAi. Dissected preparations of larval midgut from wild type (A-C), β spectrin overexpresser in fat body (*Cg*-Gal4 driven UAS-*β-Spec*^{95} ; D-F), or lipophorin knockdown in the fat body (*Cg*-Gal4 driven UAS-lipophorin RNAi (P{TRiP.HMS00265} ; G-I) were stained with lipophorin antibody. The lipophorin antibody staining pattern on the surface of the midgut in wild type (*Cg*-Gal4 parent line, A) was eliminated by either β spectrin overexpression (D) or knockdown of lipophorin (G). Anti-β_H spectrin was used as a positive staining control (B,E,H). Fat body did not exhibit β_H spectrin staining above background (arrowhead, E) but did stain strongly for lipophorin after β spectrin overexpression (arrowhead, D). Bar = 50 um.

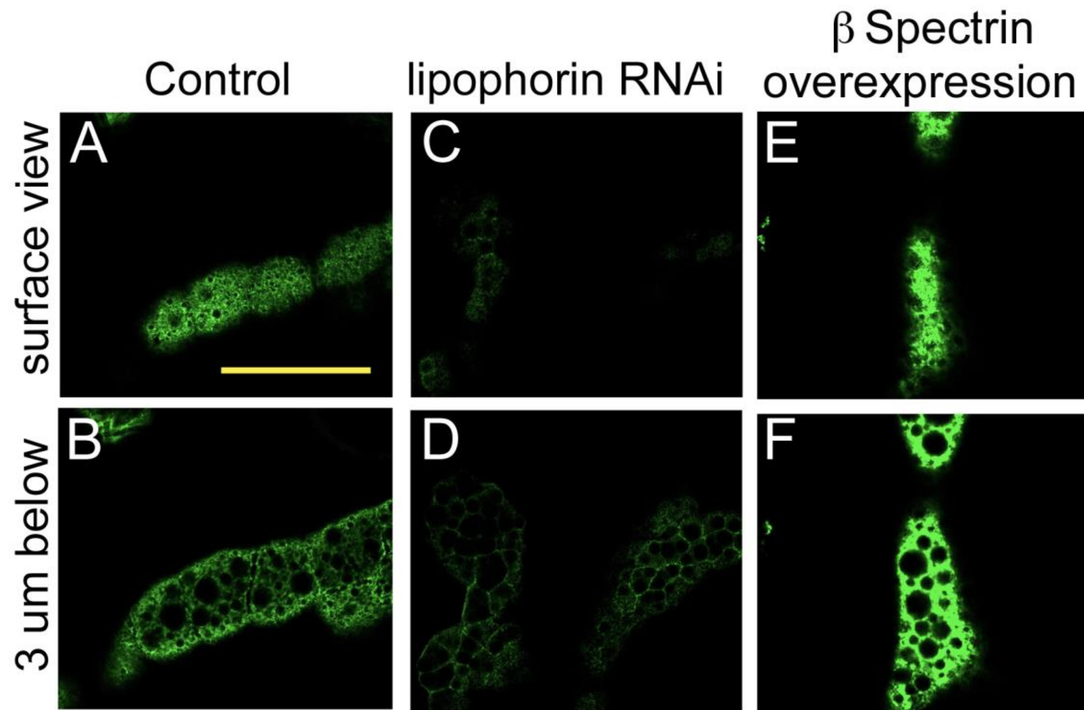

Figure S6 Effects of UAS-*β-Spec*[95] overexpression and lipophorin knockdown on lipophorin staining in larval fat body. The staining pattern in control fat body (A,B) was greatly diminished after dsRNA knockdown of lipophorin (C,D). Lipophorin staining became more intense upon β spectrin overexpression in fat body, which is consistent with a blockage of lipophorin secretion that leads to its accumulation at an abnormally high level. Bar = 50 μm.