**Supporting Figure Legends**

**Figure S1. GFP+ fast spiking basket cells and chandelier cells in layer 2/3 of G42 mouse barrel cortex, and two subtypes of Martinotti cells in layer 2/3 of GIN mouse barrel cortex.** **A-C** illustrate an example GFP+ fast spiking cell. **A** is a confocal projection image of the biocytin-stained cell. It has the morphology of a large basket cell. The two arrows in **B** point to the recorded GFP+ fast spiking cell, with the left panel showing GFP expression and the right panel showing biocytin staining (red) of the same intracellularly recorded neuron. **C** shows the cell's responses to intrasomatic current injections of 300 ms duration at amplitudes of 150 pA, 200 pA, 230 pA and 250 pA from its resting membrane potential (-56 mV). **D-F** illustrate an example GFP+ chandelier cell. The confocal projection of the biocytin-stained cell, in **D**, shows the candlestick-like axon terminals ("axon cartridges") that are characteristic of a chandelier cell. The two arrows in **E** point to the same chandelier cell, demonstrating GFP expression in the recorded neuron. **F** shows the cell's responses to intrasomatic current injections of 120 pA and 200 pA from its resting membrane potential (RMP, -52 mV, top traces), and 300 pA and 350 pA at -65 mV (bottom traces). Thin black bars in **C** and **F** indicate the current injection duration.

**G** and **J** show examples of the morphologies of the two subtypes of GFP+ Martinotti cells (SOM+/CR- Martinotti cells and SOM+/CR+ Martinotti cells), respectively. **H** and **K** show that the recorded GFP+ cells are identified as CR- or CR+ based upon immunostaining against CR, respectively. The micrographs in **H** and **K** consist of the

images of their GFP expression, biocytin staining, CR staining and the overlays. Small white boxes in **H** denote the locations of the CR- GFP+ cell; while the arrows in **K** point to the CR+ GFP+ cell. Essentially, all the GFP+ layer 2/3 cells in GIN mouse cortex are immunopostive for SOM (see Xu et al., 2006). **I** and **L** illustrate responses to intrasomatic current injections of 50 pA and 100 pA from resting membrane potentials (-51 and -45 mV) for the SOM+/CR- and SOM+/CR+ cell, respectively.

**Figure S2**. **Three subtypes of GFP+ multipolar inhibitory cells, bipolar inhibitory cells and neurogliaform cells in layer 2/3 of G30 mouse barrel cortex.**

**Multipolar inhibitory cells (A-J):** **A** shows the morphology and GFP expression in a biocytin filled irregular spiking cell. **B** and **C** show its irregular spiking patterns both from its resting membrane potential (RMP, -52 mV) (left column) and from hyperpolarized membrane potential (-65 mV) (right column) in response to current injections. **D** shows the morphology and GFP expression in a biocytin filled regular spiking cell. **E** and **F** show its regular spiking patterns both from its resting membrane potential (-52 mV) (left column) and from hyperpolarized membrane potential (-65 mV) (right column) in response to suprathreshold current injections. **G** shows the morphology and GFP expression in a biocytin filled burst spiking cell. **H** and **I** illustrate that the example cell had regular spiking patterns from its resting membrane potential (-52 mV) (left column), but it showed bursting from hyperpolarized membrane potential (-65 mV) (right column) in response to suprathreshold current injections. (Note that all the 3 example cells happened to have the same resting membrane potential of -52 mV.)

**Bipolar cells and neurogliaform cells (J-O): J** and **K** show the morphology and GFP

expression in a biocytin filled bipolar cell having a bitufted and slender dendritic field. **L** shows the cell's responses to intrasomatic current injections of 50 pA, 100 pA and 150 pA from its resting membrane potential (-57 mV), respectively. **M** and **N** show the morphology and GFP expression in a biocytin filled neurogliaform cell having dense and spider-web like axonal arbors. **O** shows the cell's responses to intrasomatic current injections of 180 pA, 200 pA and 250 pA from its resting membrane potential (-61 mV), respectively. Conventions as in **Figure S1**.

**Figure S3. Methods used for generation of input maps and data analyses.**

**Excitatory input maps and analyses (A-D): A** illustrates response traces of whole-cell voltage clamp recordings (at -65 mV, inward excitatory currents) from a Martinotti cell following photostimulation over a 9 x19 grid of 171 sites spaced 50 um apart. **B** illustrates corresponding photostimulation sites as a grid of squares, color coded according to analysis of the measured responses (see the Methods and below). (Traces in **A** correspond to only the most superficial 19 rows of squares in panel **B**). The complete map in **B** corresponds to 252 sites (9 columns x 28 rows) which were photo-stimulated sequentially in a pseudo-random pattern. Selected recordings shown at higher magnification inside the insert in panel **A** illustrate three different types of responses that were observed. In **A** and in these selected examples, the red markers below the response traces indicate the laser duration of 10 ms. As exemplified by the recording following stimulation at site "1", stimulation at sites within 50-100 μm of the recorded cell often generated large inward currents as a result of direct activation of the recorded cell. These currents often exceed 200 pA and decay slowly, over ~100 ms. Because these large

direct currents interfere with detection of smaller synaptic currents, these sites are omitted from further analysis (Dantzker and Callaway 2000; Schubert et al. 2001; Shepherd et al. 2003). Such sites are coded as black in panel **B**. Synaptic currents can be easily distinguished from direct currents based upon their latencies, amplitudes and shapes, as shown for stimulation at sites "2" and "3". At some sites close to the recorded cell, both direct responses and synaptic currents are detected (e.g. site "2"). In these cases, synaptic currents ride on top of direct currents or follow with longer latencies (e.g., see the current indicated by the arrow at site "2"). In these cases synaptic currents are measured and included in subsequent analyses (see the Methods). Finally, photostimulation at many sites does not directly activate the recorded cell and if any response is observed it includes only synaptic currents (e.g. site "3). **C** illustrates the type of input map that is used in the main presentation. Color-coded grid maps (e.g. panel **B**) are smoothed by linear interpolation to allow a more clear visual impression of the locations providing functional input to the recorded cell. These maps are derived from quantitative measurements of responses to stimulation, as illustrated in panel **A**. These maps are used for the purpose of visual display only. The recorded cell's location is indicated by the small black circle. The color scale codes evoked amplitudes in units of pA, calculated by subtracting the mean spontaneous currents from the currents measured following photostimulation during the 150 ms analysis window. Response sizes are color coded according to the color scale at the bottom of panel **C**. Black lines at the right side of panel **C** indicate cortical laminar boundaries for layers 1, 2/3, 4, 5a, 5b and 6, based upon histological alignment information. Black squares in **B** and gray squares in **C** indicate sites excluded from analysis due to interference of large direct currents (see

above). **D** illustrates all of the recording traces sorted according to those with (top, direct responses) and without direct responses (bottom, synaptic responses) and superimposed. Sites defined as having direct responses are those with an amplitude change greater than 2SD above the pre-stimulation baseline value within the 10 ms window following laser onset. The direct responses have a distinct shape (longer rise-time) and occur immediately after the laser onset. Previously published studies have demonstrated that this latency is too short for generation of action potentials and therefore no evoked synaptic responses are possible with such a short latency (Dantzker and Callaway 2000; Yoshimura and Callaway 2005). Traces with no direct response are evaluated automatically and assigned a value in pA. Traces with direct responses are individually assessed to identify and quantify any synaptic responses that can be detected. The analysis window covers the first 150 ms following the laser stimulation, and synaptic inputs are measured for both photostimulation and control trials at each stimulation site.

**Inhibitory input maps and analyses (E-G): E-G** are formatted similarly to the corresponding panels in **A-C**. Traces illustrated in panel **E** were recorded from a Martinotti cell and made during whole-cell voltage clamp at 0 mV, allowing detection of outward, inhibitory postsynaptic currents (IPSCs). During voltage clamp at 0 mV, two types of direct responses could be detected as shown in the inset at the bottom of panel **E**. These included inward direct responses (e.g. site "1"), as well as outward current responses resulting from direct presynaptic terminal activation (e.g. site "2", see the Methods), independent from somatic firing of inhibitory neurons. Sites at which direct responses prevented detection of IPSCs were excluded from further analysis (e.g. grey square in panel C). Actual IPSCs (resulting from somatic firing of inhibitory neurons at

stimulated locations) can be identified based upon their latencies and shapes (e.g., see site "3"). Similar to generating excitatory maps, the average current amplitudes over the 150 ms analysis window were measured for both photostimulation and baseline control trials at each stimulation site. Synaptic input values for each stimulation site were obtained by subtracting the average control value from the average values of the photostimulation trials. **F** and **G** are a color-coded grid map and a linearly interpolated map, respectively. The color scale for strength of inhibitory input (pA) is indicated beneath the map in panel **G**. The recorded cell's location is indicated by the white circle. To the right of panel **G**, short black lines indicate cortical laminar boundaries for layers 1, 2/3, 4, 5a, 5b and 6, based upon histological alignment.

**Figure S4. Cluster dendrograms from an unsupervised cluster analysis of the %EI (evoked input) data to distinguish clusters of laminar input patterns.**

**A.** Cluster dendrogram based on excitatory %EIs for layers 2/3, 4 and 5a for each layer 2/3 cell examined. The Y axis indicates the average within-cluster distance between each layer 2/3 cell in the data set. Each cell is identified according to its type by the labels along the X axis. Cells are further identified based on the average pattern of input for cells of their types. Cell types labeled in blue correspond to those that received excitatory input predominantly from layer 2/3; these include burst spiking (BS) and calretinin positive Martinotti cells (M+) (cf. **Figure 7**). Cell types labeled in red correspond to those that on average received strong excitatory input from both layers 2/3 and 5a, but weak input from layer 4; these cell types include chandelier (Ch), neurogliaform (NG), regular spiking (RS) and irregular spiking (IS) cells (cf. **Figure 7**). Cell types labeled black correspond to those that on average received balanced excitatory input from layers

2/3 and 4; these include calretinin negative Martinotti cells (M-), fast spiking cells (FS), bipolar cells (BC) pyramidal cells (P). Below the horizontal axis and beneath each label are individual color-codes indicating the %EI values from layers 2/3, 4, and 5a for the corresponding cell. Each color is a mixture of red, green and blue, with the RGB value corresponding to the appropriate mixture. As shown in the color look-up table, the red, green and blue intensities in each stripe represent %EI values for layers 2/3, 4 and 5a, respectively. In the look-up table, red values (i.e., layer 2/3 %EI) increase along the x axis as labeled; green values (i.e., layer 4 %EI) increase along the y axis as labeled; blue values (i.e., layer 5a %EI) go from 100 to 0 along the diagonal according to the values inside the boxes. Note that the dendrogram separates cells into three main groups. Cells in the group to the far right all have input patterns that are dominated by layer 2/3, as indicated by their red color codes. Nearly all of the cells in this group are labeled blue, indicating that they were cell types which on average also received dominant excitatory input from layer 2/3. The cell group to the far left includes cells that received strong layer 5a, as well as layer 2/3 input, as is apparent from the predominance of blue and purple color coding. Nineteen of the 24 cells that are labeled red, corresponding to the types which on average received strong input from layers 2/3 and 5a, are found in this cluster. The middle cluster corresponds to cells that received various mixtures of layer 2/3 and layer 4 input, and most of the cells in this group are labeled black indicating that they are types which on average received relatively balanced input from layers 2/3 and 4.

**B.** Cluster dendrogram based on inhibitory %EIs for layers 2/3, 4 and 5 for each layer 2/3 cell examined. Conventions are as in **A** except that cell labels are either red or black, corresponding to whether they belong to a type which on average received strong layer 4

inhibitory input (red: M+, M-, BC and FS) or weak or no layer 4 inhibitory input (black: remaining cell types).

Note that the dendrogram separates cells into two main groups. The group to the left includes cells which individually receive strong layer 4 input, and all of these cells are labeled red, indicating that they are types which also on average receive strong layer 4 input. The larger cluster, to the right, includes cells which individually receive weak or no layer 4 inhibitory input. This cluster includes all of the cells which are color coded black, indicating that they are of types which on average receive weak layer 4 input.

**Figure S5. Spatial resolution of laser-scanning photostimulation.** The numbers of action potentials evoked by photostimulation in recorded cells are plotted against the distance between the cell and the stimulation site. The distance refers to the vertical distance from the cell body. Plots show mean values for responses collected from 7 excitatory pyramidal cells and 6 inhibitory interneurons recorded from layers 2/3 and 4. For the pooled data (pyramidal cells and inhibitory neurons – both cell types), data points are represented as mean + 1 SE. As observed during previous control experiments (Dantzker and Callaway 2000; Yoshimura and Callaway 2005), focal uncaging by photostimulation results in the generation of action potentials only in neurons with cell bodies within 100 um (and usually $\leq$ 50 um) of the site of uncaging. This ensures that most action potentials occur from direct stimulation near a neuron's soma, not from stimulation of distal dendrites or synaptic activation via distant neurons.