POINTS OF VIEW

# Pathways

Apply visual grouping principles to add clarity to information flow in pathway diagrams.

Pathway diagrams describe the connectivity and flow of information in biological systems. Remarkably similar representations can depict everything from cell-signaling pathways to global ecological networks.

Pathways are network diagrams in which molecules, cells or species are represented by nodes and their relationships by edges. Pathway diagrams benefit from strategies used to display networks[1], but additional requirements must also be met. First, they must clearly depict patterns in connectivity—the flow of information through the pathway, encoded by the direction of the edges, is often the primary purpose of the diagram. Second, both direct and indirect relationships need to be clear for one to understand the pathway as a whole. Encodings that remove indirect relationships, such as adjacency matrices, are therefore inadequate.

One can use visual grouping to help create a hierarchy for the flow of information in a pathway layout[2] and clear alignment to emphasize node relationships. Edges should connect to a fixed number of points on node shapes (**Fig. 1a**). Basic arrowheads should be used; unnecessary stylizing or stretching of arrows should be avoided. Edge angles should be limited to multiples of 30° or 45°, and curved edges can be drawn easily using circular guides (**Fig. 1b**). We use 0.5-pt lines for edges and equilateral-triangle arrowheads with sides 2.5 pt long[3].

Conventionally, we expect information to flow left to right and top to bottom. Diverging from this standard, as well as introducing asymmetry in the layout, can emphasize differences but should be done sparingly and only when it adds to the reader's understanding. Edges that loop back to upstream nodes should flow clockwise (**Fig. 1b**).

Placing nodes on a grid assists eye movement across the figure (**Fig. 1c**). Horizontal alignment of nodes emphasizes the flow of information through a pathway, whereas radial alignment highlights source nodes (**Fig. 1d**). Local deviation from the grid pattern may be necessary to avoid crossing of edges or collisions between arrowheads (**Fig. 1c**).

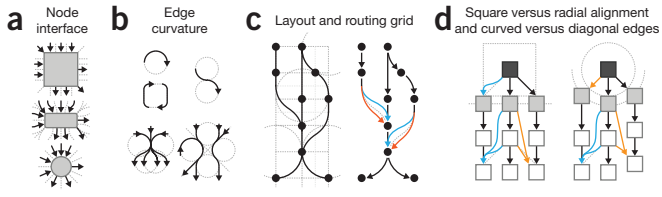


**Figure 1** | Rectilinear and curved grids provide overall consistency. (**a**) Fix the number and position of connector points and distribute edges symmetrically in quadrants. (**b**) Create consistent edge curvature with grids based on circles and rounded rectangles. (**c**) Nodes positioned on a square grid with circular edge curvature. Connecting adjacent edges with overlapping arrowheads can be locally adjusted from their circular path (dotted line) by vertical stretching (orange) or shrinking (blue) of the edges. (**d**) Neighbors (gray) of a source node (dark gray) can be aligned horizontally (left) or radially (right). Arrow length can be altered in radial alignment to create groups. Curved (blue) and diagonal (orange) edges are locally adjusted from a 45° guide to avoid overlap between arrowheads.

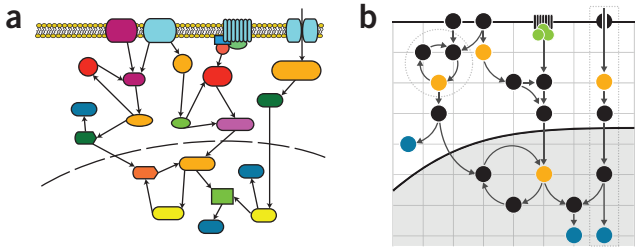


**Figure 2** | Creating a clear pathway diagram using a grid and directional information flow. (**a**) Example of a pathway diagram with unorganized information flow, unnecessary visual detail (e.g., membrane lipids) and no visual continuity along pathways. The dashed line represents the cell nucleus. (**b**) Pathway from **a** with redundant visual encodings removed and main points emphasized by visual grouping. Color and shape variations have been removed except for those highlighting a molecule of interest (orange), the products of the pathway (blue) and a membrane protein complex (green). Gray lines are layout guides and would not be included in the final figure; the solid lines provide a grid, and the dotted lines highlight the strong grouping effect of visual connection (circular paths) and proximity.

Strong relationships between pathway components can be illustrated using connection and enclosure[4]. Edges act to group nodes via connection, whereas enclosure can be used to group nodes in shared compartments, such as the nucleus (**Fig. 2**).

Associating nodes through similarity (e.g., color or shape) or proximity can highlight parts of a pathway without interrupting the groupings created by connection and enclosure. For similarity groups to be effective, unnecessary variation in the color or shape of nodes should be avoided, except when used to highlight nodes (**Fig. 2b**). Proximity grouping can be achieved with negative space—an empty row or column on the grid around the group adds visual emphasis (**Fig. 2b**). Differences can be identified with labels or with an unambiguous shape associated with a specific protein class. For example, in **Figure 2b** we show the seven transmembrane domains of the GPCR, and the G protein as a green complex.

The use of grouping in pathway diagrams can provide alternate visual entry points. In a busy pathway, it can be difficult to work from start to finish through all possible paths. When important node subtypes are easily identifiable, a pathway diagram can be examined from several directions instead of strictly serially.

Adding labels to nodes is often a challenge—names of genes and protein complexes can be long, but altering node shapes to fit labels dilutes grouping effects (**Fig. 2a**). It is a good idea to choose node shapes that accommodate the longest label, or abbreviate names. Keep node colors desaturated to avoid loss of contrast with the text, and avoid visual garnishes such as gradients and drop shadows.

Next month we will explore how to encode multiple variables in pathway diagrams by taking a closer look at neural circuits.

**COMPETING FINANCIAL INTERESTS**
The authors declare no competing financial interests.

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