Supplemental Movie 1. Proliferation of HT-1080 fibrosarcoma cells in a thiol-ene hydrogel. HT-1080 cell divisions were common during the 6 hour time course of a typical experiment, with cells migrating away from one another, demonstrating that thiol-ene hydrogels are permissive towards both proliferation and migration. Note that the HT-1080 cell that migrates to the right undergoes a second cell division. For this movie, HT-1080s were seeded in a thiol-ene hydrogel with 1000 μM RGDS as described in Experimental.

Supplemental Movie 2. Migration of dermal fibroblasts in a thiol-ene hydrogel. Fibroblasts migrate in thiol-ene hydrogels through mostly random movements characterized by spread morphologies with multiple protrusions. For this movie, fibroblasts were seeded in a thiol-ene hydrogel with 1000 μM RGDS as described in Experimental.

Supplemental Movie 3. Migration of HT-1080s in a thiol-ene hydrogel. HT-1080s migrate in thiol-ene hydrogels in a highly persistent manner and are characterized by a much more rounded morphology than was observed for fibroblasts, with a leading edge protrusion that typically defines the direction of migration. For this movie, HT-1080s were seeded in a thiol-ene hydrogel with 1000 μM RGDS as described in Experimental.

Supplemental Movie 4. A highly persistent HT-1080 migrating in a thiol-ene hydrogel. HT-1080s often migrated with only limited change of direction during the 6 hour time course of observation, leading to cells traveling through long, highly persistent paths. This movie also highlights the rounded cell body and single leading edge protrusion that was common for HT-1080s seeded in thiol-ene hydrogels. For this movie, HT-1080s were seeded in a thiol-ene hydrogel with 1000 μM RGDS as described in Experimental.

Supplemental Movie 5. Examples of constriction rings forming during HT-1080 migration in a thiol-ene hydrogel. HT-1080s forming apparent constriction rings are highlighted with a box. A constriction ring is a feature that allows cells to move through spaces that are smaller than the cell body and has previously been observed for cells migrating through an amoeboid mechanism (for example, see References 12 and 38). For this movie, HT-1080s were seeded in a thiol-ene hydrogel with 1000 μM RGDS as described in Experimental.

Supplemental Movie 6. HT-1080 migration in collagen. HT-1080s migrating in collagen appear more elongated than what was observed in thiol-ene hydrogels, similar to previous studies (for example, see References 12, 15, and 17), and demonstrating that the specific strain used here was not predisposed towards highly rounded morphologies. For this movie, HT-1080s were seeded in 3 mg/mL bovine collagen (PurCol, Advanced Biomatrix).

Supplemental Movie 7. 2D Fibroblast migration on tissue culture polystyrene. Fibroblasts adopted a typical spread, fibroblastic morphology on tissue culture polystyrene. Migration was limited for fibroblasts seeded in 2D.

Supplemental Movie 8. 2D HT-1080 migration on tissue culture polystyrene. HT-1080 morphology appears very different than fibroblasts, even in 2D (For fibroblast migration in 2D, see Supplemental Movie 7). While many HT-1080s migrated through an apparent mesenchymal mechanism, others appeared more rounded with a single, flattened leading edge protrusion. Rounded HT-1080s appear to glide along the surface rather than exhibiting mesenchymal-like process extensions and detachments, which is similar to what has been observed for monocytes (see Reference 50).