

Time-lapse projection of real time video reveals substantial detail of *C. elegans* locomotion on agarose and swimming in liquid culture

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E. coli lawns on agarose in Petri dishes and liquid culture in 96- or 384 multi-well plates are standard cultures for *C. elegans*. Wild type, mutant and drug-induced locomotion can be scored on agarose in great detail, because of the relatively low speed and of the subtle effects that can be discriminated in a 2 dimensional plane. In liquid culture, swimming occurs in 3 dimensions, is more vigorous and therefore less amenable to subtle microscopic scoring. In both cases, viewing and reviewing many nematodes of a culture is required to determine exactly if and with what penetration a phenotypic behavior differs from wild type or vice versa. We aimed at developing software that can reduce the hands-on time needed to evaluate locomotion in genetic or compound screening, using video and automated image analysis.

Time-lapse projection of locomotion on agarose. Microscopic video sequences of wild type untreated and treated *C. elegans*, were deconvoluted into stacks of images and in each image the nematode was identified by specific image segmentation. The derived nematode binary images were then skeletonized to a line of pixels reflecting the central body axis and projected on each other with different time lapse intervals to create a single 'hairy rope' image that reflects the key features of the video sequence. These images displayed clearly distinct features of wild type and acetyl cholinesterase-inhibited locomotion, which are in line with the phenotype of the *ace-1*, *ace-2* double mutant.

Swimming in liquid culture. Nematodes in 96-well plates were treated with a dose range of toxic compounds and automatically imaged using the MIAS-2 microscopic reader. Two consecutive images were taken with an interval of 200 milliseconds and locomotion was scored by subtracting two images and projecting the segmented difference on top of one image in pseudo color. This imaging script proved to be very robust in highly variable conditions, and was successfully applied in LD₅₀ compound testing with the nematode.

We propose 'hairy rope' and motility difference images as a tool to evaluate behavior wild type, mutant or drug-induced changes in behavior.

Abstract Data View

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Abstract:

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