to transfer flies from one vial to another without losing some of them. This suggests a question as to whether the material properties of the cuticle in these species that are necessary to support their physical dynamism produces an effect in imaging contrast, and that it may be interesting to study insect biomechanics with MRI.

Successful injection of embryos with contrast agent would be an exciting step forward. The possibility of injection of embryos with MRI contrast agents was not fully explored here, and offers fertile ground for future experiments. In general, the primary difficulty facing contrast agent experiments in model organisms is the challenge of how to actually get the contrast reagent to the interior of cells so they can be used as indicators of physiological states such as pH, calcium ion measurement and gene expression can be studied *in vivo*. *Drosophila*'s syncitial development has long been utilized for transgenic and RNAi methods, and could present a special opportunity for use of MRI contrast agents in a genetic model organism, due to the potential to load cells with contrast agent prior to cellularization. If an adequate concentration of contrast reagent is retained in the cells following subsequent divisions, the fly will have intracellular contrast agent in all its cells or only within specific cell lineages. In our preliminary attempt to inject embryos, no optimization was done to the formulation of the solution, and it was beyond the focus of our current effort to pursue the method further. Improvement of the buffer solution used for injection could lead to the development of an intracellular *in vivo* contrast agent system for the fly, a very exciting possibility. Such a method would provide an excellent opportunity for utilizing existing MRI contrast agents and encouraging the development of new varieties of contrast agents for molecular genetic studies.

Imaging with other nuclei:

Carbon imaging (C-13) on an untreated sample was attempted, but did not produce a successful image. Fluorine presents a stronger signal than carbon, and is not normally found in organisms naturally, so could be an interesting direction for imaging labeled molecules. Notably, bacteria can produce fluorinated proteins from fluorine-tagged amino acids, and thus might be used to synthesize *in vivo* labeling agents for use in fluorine imaging.

Acknowledgments: This work was enabled by cooperation and support from Professors Joseph Puglisi, Structural Biology Chair and SMRL Director, and Michael Moseley, Lucas Center Radiology Science Laboratory, in the Stanford School of Medicine. Key assistance was provided by Matthew Fish (embryo injection), and John Sack & Richard Aldrich, and immeasurable support was provided by Varian Inc. to promote and support the advancement of this exciting technology.

Economical high-throughput DNA extraction procedure in 96-well format for *Drosophila* tissue.

Holleley, Clare E. School of Biological, Earth and Environmental Sciences, University of New South Wales, Sydney, NSW 2052, Australia. E-mail: c.holleley@unsw.edu.au.

One of the major benefits of working with *Drosophila* is the ease of achieving highly replicated and statistically powerful experiments. However in molecular studies, the time consuming process of single-tube DNA extraction can create a work-flow bottle-neck. We have modified the standard salting-out DNA extraction method so that it can be conducted in 96-well PCR plates, dramatically increasing the number of extractions that can be performed. The salting-out method is

often preferable to column based extraction methods, because you can manipulate the final volume in which you re-hydrate the purified DNA. Thus it is ideal for species with low expected DNA yields. Also, salting-out DNA extraction methods are usually much less expensive than column based extraction methods. We modified the manufacturer's instructions provided in the Gentra PUREGENE® DNA purification kit (Progenz Ltd, Australia) for use in 96-well plates. However the methods described here can also be applied non-commercial salting-out procedures (Aljanabi and Martinez, 1997; Sunnucks and Hales, 1996).

We first overcame the time consuming process of individually macerating each *Drosophila* tissue sample using a stainless steel 96-well phage colony replicator tool with 1.6 mm round-ended Phage Picker+ pins (Genetix, Millennium Science, Australia). The pins of this commercially available instrument fit exactly into the wells of a standard 96-well PCR plate. Using this instrument, we could macerate 96 tissue samples simultaneously in the chilled cell lysis solution (Tris, EDTA, SDS) by grinding for a minimum of 10 minutes. After macerating each *Drosophila* in 100µl of cell lysis solution, we added 0.5µl of Proteinase K (20µg/ml) to each well. We then sealed the plate, mixed the samples and incubated the samples overnight at 55˚C in a PCR machine.

The following morning, the samples were cooled to room temperature and 50µl protein precipitation solution (ammonium acetate) was added to the cell lysate and vortexed vigorously for at high speed for 20 seconds. Samples were then centrifuged at 4100 rpm for 12 minutes to pellet cell debris. Immediately after centrifugation we *carefully* removed approximately 100µl of supernatant, without disturbing or sucking up any of the pellet using multi-channel pipette. This step required careful observation of marker lines on the pipette tips to gauge depth, and we found that elevating the plate aided visibility. If the pellet is disturbed, re-centrifuge the samples. We then added the supernatant to 100µl of room temperature 100% isopropanol in a new 96-well PCR plate. We sealed the plate, inverted the plate 50 times to mix and then centrifuged at 4100rpm for 12 minutes to pellet the DNA. We then removed the supernatant and washed the DNA pellet by adding 150µl of 70% undenatured ethanol and inverting 10 times. The plate was then centrifuged again for 12 minutes. It was essential to remove all ethanol from the DNA pellets prior to rehydration. Thus if ethanol was still visible in the wells after draining, we inverted the plate on several kim-wipes and centrifuged upside down at 200 rpm for 30 seconds. We then let the plate air-dry for 10-15 minutes. After all visible ethanol had been removed from the wells, we re-hydrated the DNA in the desired volume of hydration solution (TE Buffer: 10mM Tris-HCl, 1mM EDTA, pH 7.0-8.0) or RNase-free water. Finally we re-hydrated the DNA pellet by incubating sample at 65˚C for 1 hour or incubating at room temperature overnight, tapping periodically to disperse DNA. For a single fly, the observed DNA yield ranges from 35ng – 10µg with a mean concentration of 172.4 ng/µl ± 21.3 SE (N = 189) when re-hydrated in 20µl. The failure rate is low (2.1%) and is usually due to a cracked well during excessively vigorous maceration.

References: Aljanabi, S.M., and I. Martinez 1997, Nucleic Acids Res. 25: 4692-4693; Sunnucks, P., and D.F. Hales 1996, Molec. Biol. Evol. 13: 510-524.

Call for Papers

Submissions to Drosophila Information Service are welcome at any time. The annual issue now contains articles submitted during the calendar year of issue. Typically, we would like to have submissions by 15 December to insure their inclusion in the regular annual issue. but articles can be accepted for this volume until 31 December. Details are given in the Guide to Authors or on the DIS web site: www.ou.edu/journals/dis.