became reduced by 50 percent after implementing our technique. Thus if one has to scan the entire second chromosome using the DrosDel deficiency kit that has 109 deficiency stocks, our method can reduce the workload by 43 percent, meaning that one only needs to use 62 deficiency stocks for the first round of crosses.

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Drosophila Proteome Atlas.

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Since the first report of the *Drosophila* genome project in 2000 (Adams *et al.*, 2000), DNA sequence information became a valuable asset to research projects using *Drosophila* as a model organism. However, the knowledge of *Drosophila* proteins that are expressed and thereby manifest the function of the genome is far behind the success of its genetics and genomics counterparts. We would like to draw the attention of *Drosophila* researchers to our effort to provide information on proteins based on proteomics inquiries in which proteins are the immediate subject matter. In our recent paper (Takemori *et al.*, 2007) we reported 1) that proteomics profiling of *Drosophila* compound eyes in comparison to brain is a powerful tool to investigate post-translational modification of proteins using tissue-specific calmodulin methylation as an example, and 2) that such proteome information will be a useful asset to the *Drosophila* community. In our protocol, microdissection of the compound eyes and brain from dehydrated tissues provides clean and sufficient materials for analysis on a two-dimensional (2-D) gel electrophoresis (Matsumoto *et al.*, 1982; Matsumoto and Pak, 1984). Furthermore, a multi-stage mass spectrometric analysis of a 2-D gel spot allows us to determine the structure of the modified amino acids at a microscale (at ~100 fmol levels) (Takemori *et al.*, 2006). With a belief that the information obtained in this work and that to be obtained in our future work will benefit other *Drosophila* researchers, we initiated an open access protein database "*Drosophila Proteome Atlas* (DPA)" posted at The University of Oklahoma (Drosophila Information Service) and at Kyoto Institute of Technology (Drosophila Genetic Resource

Center): http://www.ou.edu/journals/dis/ProteomicsDatabase/Proteomics_Home.htm and http://www.dgrc.kit.ac.jp/~jdd/proteome/Proteomics_Home.htm, respectively.

In constructing DPA we start from protein maps displayed on two-dimensional (2-D) gel electrophoresis. Protein is identified by peptide mass fingerprinting followed by the confirmation by MS/MS (Matsumoto *et al.*, 2005). The 2-D gel electrophoresis renders a map of proteins that exist in the sample of interest, giving visually comprehensible images carrying rich information on proteome, including information on post-translational modification of proteins and the quantity of expressed proteins, neither of which can be elucidated from genome information *per se* (Matsumoto and Komori, 2000; Matsumoto *et al.*, 2005). In the long-standing effort to build a biological project from a physiological phenomenon one of the authors started observing the light-induced post-translational modifications of proteins in the compound eyes of *Drosophila in vivo* (Matsumoto *et al.*, 1982; Matsumoto and Pak, 1984). Because of the lack of technical availability it took several years for us to clone the gene that encodes the protein of interest (Yamada *et al.*, 1990). Meanwhile, two key ingredients in proteomics, *i.e.*, genome information and modern mass spectrometry, emerged in different areas of science and technology and, finally, merged as a new interdisciplinary area of bioscience (Matsumoto *et al.*, 1999). Although we started the Atlas from the compound eye in reference to brain, we plan to add more maps of other tissues and organs of *Drosophila* in our future effort, thereby making the *Drosophila Proteome Atlas* increasingly useful to other researchers. It is our belief that proteomics information will add a new dimension to research projects using *Drosophila* and accelerate their progress.

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Targeted replacement of *piggyBac* transposable element.

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Since Rubin and Spradling first developed the use of *P*-elements for transgenesis (Rubin and Spradling, 1982), the *P*-element has become an indispensable tool. Gene tagging, enhancer trapping, gene disruption, chromosome engineering, and inducible gene expression have taken advantage of the mobility of *P*-elements (Ryder and Russel, 2003). However, a *P*-element also has drawbacks, such as its strong bias in insertion sites. For reasons that are not entirely clear, it has a preference for some chromosomal locations (hot spots), but avoids others (cold spots) (Spradling *et al*., 1995).

The *piggyBac* element was discovered in the cabbage looper moth and has proven to be an efficient vector for the transformation of many insect species as well as human and mouse cells (Ding *et al.*, 2005). Because *piggyBac* has a different insertion site preference than that of a *P*-element, it has been used in several large scale gene disruption projects as a complement to *P*-elements (Hacker *et al*., 2003; Bellen *et al*., 2004; Thibault *et al*., 2004).

After a *P*-element excises from a chromosomal location, another *P*-element from a second site frequently transposes into the site of the excised *P*-element. This process is called "*P*-element replacement" (Gonzy-Treboul *et al*., 1995) or "targeted transposition" (Heslip and Hodgetts, 1994; Sepp and Auld, 1999). To date, however, similar replacement between *piggyBac* transposons has not been reported. We tested whether a *piggyBac* transposon, PB[y^+] on the 2nd chromosome (Bloomington stock ID16249, originally created by Ring and Garza) (Bellen *et al*., 2004), can efficiently transpose into the site of another *piggyBac* element, RB[w^+] e03162 (from Exelixis Collection at the Harvard Medical School), which is located in the second intron of the *slowpoke* gene (Thibault *et al*., 2004). We remobilized PB[y^+] in the presence of RB[w^+] e03162 using the constitutive {α-1 tubulin:*piggyBac* Transposase} transgene (Parks, 2004). Four out of fifteen crosses produced one or more potential replacements based on the marker phenotype. Two of those lines were molecularly characterized by PCR using one primer specific to PB[y^+] and the other from the *slowpoke* genomic region, followed by DNA sequencing. We found that in both cases, the replacement appears to be precise, leaving the local genomic DNA outside the transposon unchanged.

Since thousands of *piggyBac* insertions are widely distributed in the *Drosophila* genome and are available from stock centers (Bellen *et al.*, 2004; Thibault *et al*., 2004), the efficient targeted replacement of *piggyBac* opens up many possibilities for precise genetic manipulations and chromosome engineering. For example, a UAS containing *piggyBac* element could be placed at a desirable genomic position that harbors an existing *piggyBac* element without the UAS sequence.