

ABSTRACT

LOGHIN, ARINA. Who is an Actor? Analyzing Agency in a Lab's Social World. (Under the direction of Nora Haenn).

The notion of agency has a long history in the theorization of laboratory work. This study contributes to the conversation about agency by bringing insights from the ways in which scientists interact with the material and biological tools in the genetics laboratory. Qualitative data was obtained through participant observation and interviews in a *Drosophila melanogaster* (fruit fly) laboratory of North Carolina State University.

The focus of this research is the aspects about which researchers are constantly mindful when performing laboratory tasks. The study follows *Drosophila*'s transformation in the lab from living model organism to DNA/RNA samples to a series of data points, and eventually data analyses. This transformation occurs in three stages, which are not consecutive. In the fly work stage, scientists conduct assays on fruit flies to test their behavior or measure physical traits. In the molecular stage, scientists conduct molecular protocols using flies' DNA or RNA. In the data analysis stage, researchers conduct statistical analyses, and use GWAS Pipeline to make correlations between the phenotype and genotype of fruit flies.

This study emphasizes the ambivalence that the scientists in the *Drosophila* lab have towards their tools' agency. It argues for the importance of situating agency in the social community of the laboratory studied. Rheinberger's concept of instrument-experimental object interface and Knorr-Cetina's concept of indeterminacy help frame the role of the scientist as primary decision maker in the *Drosophila* lab. Furthermore, the notion of sociality brings forth the significance of the social context of the laboratory.

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Who is an Actor? Analyzing Agency in a Lab's Social World

by
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DEDICATION

To the scientists in the *Drosophila* lab.

BIOGRAPHY

The author of this thesis was born in Romania. Upon finishing High School she enrolled at the American University in Bulgaria where she studied history, fine arts and anthropology for three semesters. A semester abroad within the Erasmus Programme at the University of Pécs in Hungary followed. While in Hungary, she transferred to University College Maastricht in the Netherlands. She graduated with a B.A. in Liberal Arts in 2011. She worked for one year at University College Maastricht on an educational project under a Leading in Learning grant provided by Maastricht University. In 2012 she received a Fulbright scholarship and she enrolled as a master student in Cultural Anthropology at North Carolina State University.

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CHAPTER 1 INTRODUCTION

Scientific facts are produced in part through experiments. At the core of any experimental system are the equipment, the experimental objects (i.e. organisms, substances) and the scientists. To get a better grasp of how knowledge is produced, it is instrumental to understand the relations between these elements in the experimental process. Analyzing how scientists interact with their equipment and experimental objects is important, because the scientists coordinate the output presented to wider audiences. To understand laboratory dynamics, I conducted ethnographic fieldwork for two months in a *Drosophila melanogaster* (fruit fly) laboratory of North Carolina State University. At the same time, I had the opportunity to experience fly work by conducting an individual project using flies in that lab.

My study aimed to understand *the interactions between scientists and the tools in the lab during Drosophila's transformation* from model organism to statistical model. To accomplish this aim I elucidate *the research stages in which Drosophila transforms physically in the lab*. Furthermore, I observe *the kinds of concerns scientists have when they use their tools* throughout the research process. “Concerns” refer to aspects about which scientists in the lab are constantly mindful and recurrent issues with instruments that they need to prevent.

As such, my inquiry focuses on three different types of interactions that correspond to three core stages of the research process in which scientists systematically use lab equipment: the first is between scientists, *Drosophila* and the equipment used for observing the flies; the second is between scientists and the equipment used for molecular work on genes of interest;

the third is between scientists and their statistical software and analyses of *Drosophila* gene-behavior interaction. The employees in the lab who participated in this study included undergraduate assistants, technicians, PhD students, postdocs, senior researchers. I interchangeably refer to them throughout the text as “scientists”, “researchers”, “experimenters” or “lab staff”.

The concerns reveal the limitations of assuming the agency of all things present in a social environment prior to studying it, a perspective advocated by non-anthropocentric approaches to agency (Latour 1988, 2005; Knappett & Malafouris 2008). I exemplify several conceptualizations of agency which assume that agency characterizes both humans and non-humans in the lab. For some, agency is intrinsic to all objects and organisms (Rothbart 2007). Others explain how the components of a network impact one another (Latour 1988, 2005; Callon 1986). Yet others assume agency is something conditional, based on biological or cultural needs, or delegated by humans unto non-humans (Kaptelinin and Nardi 2006). In the lab studied, these conceptualizations are only faintly illustrated in the way the researchers see the agency of objects and flies.

Consequently, I draw on notions of indeterminacy (Knorr-Cetina 1981) and instrument-experimental object interface (Rheinberger 2010) to argue for the contextualized character of agency. Indeterminacy refers to selecting instruments, theories and processes in the contingent context of the laboratory (cf. Kleinman 2003). The instrument-experimental object interface refers to the intersection between the instrument and the experimental objects (model organism, tissue samples etc.) which inform the craft and knowledge of the scientist.

These theories help to position the scientist as primary agent in the lab. Additionally, the norms of sociality (meaning-making and relationships in a social community) serve to nuance the relations between scientists and their tools in the cultural context of the lab.

Following the scientists, their tools and *Drosophila*'s transformation in the lab from model organism to DNA/RNA sample, to data points and statistical analyses, I saw how scientists constantly affirm their own agency. At the same time, the scientists in question displayed ambivalence with respect to attributing agency to instruments and fruit flies in the different stages of research. The following pages focus on scientists' own ideas of agency as these emerge from practice and speech to illustrate the variable quality of agency as they understand it and, thus, the importance of contextualizing agency in specific ethnographic circumstances.

I give special attention to ideas of agency in light of the complexity of tools use in the biology laboratory. Here, I do not consider the rhetorical dimension (Latour and Woolgar 1979) of the notion of tool (i.e. the role of tools in the wider social circuit of the knowledge produced in the lab). I use the term "tool" for all the material and biological resources in the fly lab that scientists manipulate to rear and experiment with flies, including resources in the molecular lab, the computers and data analysis software. In presenting results, I mainly use "instrument" to refer to material tools, to underscore the differences in scientists' approaches to material equipment as opposed to flies. I will come back to this point in the *Discussion*.

Theorization of laboratory work on model organisms has largely employed historical or philosophical perspectives (Rheinberger 1997; Rothbart 2007; Creager, Lunbeck & Wise

2007). By exploring the relationships among lab staff, flies and lab equipment I uncover various ways scientists position themselves with respect to their tools during each stage of the research process. In relation to their material tools scientists are mindful about avoiding cross-contamination, consistency of manipulation, proper positioning and handling of instruments and the timing of experiments. Scientists also encounter equipment breakdowns. When talking about their concerns with respect to material tools sometimes scientists use language that suggests objects have an agency of their own. However, when they physically manipulate tools their behavior suggests scientists only consider themselves as agents. With respect to flies, researchers' practice and speech mostly acknowledge flies' agency, as they are living organisms who have particular needs and the ability to act. However, scientists' speech also suggests flies are tools that they control. In this context, flies become non-agents. Fly DNA is treated only as non-agent. Similarly, flies as data points are only objects, non-agents in researchers' view.

The findings matter within the debate around the nature of agency. By noting scientists' ambivalence we learn that the conceptualizations of agency demand greater specificity, as well as consideration of norms of sociality. Scientists are not thinking in complex terms about the agency of their tools. They are thinking however about the dynamic role of objects in their social world. For them, objects become agents when they exert to an extent the ability to act (such as software or machines), or oppose resistance. We can define these interactions using sociality, as this concept brings forth the role of scientists in creating meaning in their social world.

Drosophila melanogaster has been a powerful model organism in the genetics laboratory since the inception of laboratory genetics research (Kohler 1994; Weber 2007). In spite of its historical importance, many contemporary publics know little about research on *Drosophila*; nor do they understand the relevance of using the insect as a model organism in genetics. By investigating the trajectory of the fly through the different stages of a research project, and describing the tools and processes that scientists use, I further aim to improve the reader's knowledge of the contemporary genetics laboratory.

Below, I present an overview of the literature that informed this study. I then describe the data collection methods, followed by a presentation of the physical organization of the laboratory. The three main parts of laboratory work include: fly work, involving behavioral experiments; molecular work using molecular protocols; data analysis, using statistical software. The bulk of the thesis employs ethnography to understand scientists' changing notions of agency across these settings. In the discussion, I address the patterns in the physical transformation of the fly, as well as insights from researchers' ambivalence towards the agency of flies and equipment. I conclude by elucidating the contextual nature of agency and its potential broader implications.

CHAPTER 2 LITERATURE REVIEW

Critical analysis of the modern scientific process has a fairly short history. Science and Technology Studies, which grew in the Cold War period, offered works in the history and philosophy of science, such as the prominent *The Structure of Scientific Revolutions* by Thomas Kuhn. Studies of laboratory life began in the 1970s with the emergence of the Sociology of Scientific Knowledge (SSK). This new school of thought emphasized science as a product of social interactions.

With their ethnographic fieldwork at a research institute, the first of the kind in the field, Steve Woolgar and Bruno Latour (1979) introduced a novel conceptual system to understand knowledge production in the laboratory. Woolgar and Latour analyzed how people in a neuroendocrinology laboratory used equipment and conducted experiments. They asserted that the equipment in the lab acts as a source of inscriptions which the scientists interpret and then put together as texts.

Other social science researchers of laboratory practices joined Latour and Woolgar. Knorr-Cetina (1981) emphasized the discrepancies between the end product of a research project and the actual research process. For her, scientific work involves selecting instruments, theories and processes in the contingent context of the laboratory, a place characterized by particular instruments, people and relationships between these (cf. Kleinman 2003). For Knorr-Cetina this contingency means scientists always work in situations indeterminacy in which their decisions and actions depend on their interpretations of

particular situations within the laboratory, including the resources and equipment present there (Freudenthal 1984).

A relevant aspect for the present study of the theory on laboratory work has been the analysis of the *physical* interactions between humans and the objects in the lab. The history of empirical processes of knowledge production highlights the importance of the interactions between scientists, experimental objects and laboratory equipment. Rheinberger depicts experiments as complex systems driven by the interaction between instrumentation and model organisms in biological laboratories (2010; 1997).

Latour and Woolgar (1979) and Rheinberger (2010) analyze laboratories from different perspectives. As such, they present differently the scope of the functions lab equipment. Latour and Woolgar (1979) observe the route of a brain sample from one machine to another and analyze the processes that take place in relation to text production. Importantly, Latour and Woolgar analyze equipment from a broader social perspective; the equipment is embedded in a larger circuit of literary production which fulfills a social function of disseminating knowledge. Later, Latour thought that humans and non-humans in the lab form networks of actors (or actants, as he calls them). According to Latour (1987: 68) an instrument, or inscription device, is a set-up of any size, shape and cost that provides a visual display of a scientific text. Instruments have rhetorical power and play an important role in demonstrating the scientists' access to nature (Latour 1987). This can be seen especially in controversies.

In contrast, in his philosophical analysis of laboratory equipment and model organisms, Rheinberger (2010) focuses on the space of the lab and its internal processes of producing reactions and interactions between machines, objects of inquiry and scientists. He elaborates on the materiality of research, by avoiding the view that science-making is necessarily theory-laden as philosophers of science proclaim. For Rheinberger, the analysis starts from the role a piece of equipment plays in the experimental system. He claims that instruments in the biological sciences produce “very diversely configured intersections between themselves and the objects they are used to investigate” (Rheinberger 2010: 217). He refers to such intersections as *instrument-experimental object interface*, with which the experimenter interacts through observation and handling. The investigative value and productivity, as well as the epistemic meaning, of an instrument lies at this intersection as observed in particular experimental systems (Rheinberger 2010). In this way, an instrument demonstrates its utility and elicits the association of certain meanings with its use, only through the observed effects of its technical relation with experimental objects. By demonstrating utility and eliciting meaning, the instrument-experimental object interface informs the experimenter’s actions and craft. In this way, Rheinbrger’s ideas help premise affording priority to scientists’ agency.

The theoretical frameworks presented above are linked to the way agency has been conceptualized in the lab, as well as more broadly, and how it could be conceptualized for a more substantive understanding of lab interactions. The debate around the meaning and use of agency in the humanities and social sciences is complex (Knappett & Malafouris, 2008),

encompassing a spectrum of conceptualizations ranging from anthropocentric approaches (affording agency only to humans) (Giddens and Pierson 1998) to non-anthropocentric approaches (recognizing that all humans and non-humans have agency) (Latour 1988, 1999, 2005; Knappett & Malafouris, 2008). Some anthropological writings (Mauss 1954, Appadurai 1986) suggest a humanistic determination of agency, which is different than anthropocentric. The humanistic determination emphasizes the way that throughout the evolution of cultures, humans have endowed objects, and non-human beings with symbolic powers to act, thus animating them (Nurrit-Bird 1999; Franke 2010). Below I critique the non-anthropocentric perspectives of agency, arguing for the importance of situating agency in the specific cultural context studied.

Material agency in the lab has a long history in the Western development of the philosophy of science and is connected to a conceptualization of the living world as “a series of smaller machines embedded in larger ones” (Rothbart 2007, 69). This view dates back to the 17th century when natural philosopher Robert Hooke, who developed natural microscopy, maintained that “[e]very tool is an agent for change, that is, a source of movement that is responsible for certain occurrences” (see Rothbart 2007, 69). Hooke arrived to this conclusion by observing organisms under the microscope where he said they act as “an agent for change, as well as a reagent subjected to influences by other bodies. In this respect, a specimen is knowable by its capacities to produce, create, and generate detectable events” (Rothbart 2007, 108). The point here is that all have intrinsic properties for action. Minute particles, or machines, move against one another facilitating these properties.

Hooke's idea of agency seems grounded in a rational approach to the nature of objects. For the purposes of this thesis, it is important to note that this early example already premises the scientist in conceptualizing the nature of agency in scientific work.

Latour's Actor Network Theory (ANT) is an influential framework in the social science of laboratory research (Latour 1988, 2005; Callon 1986). Classical ANT provided a methodology for thinking about the impact of equipment and experimental objects on scientists' behavior and for assembling the connections between these actors. In Latour's characterization, scientists represent all the objects they use in the laboratory; as such, they are spokespersons for the symbols instruments produce (Latour 1987: 84). At the same time, ANT posits that lab equipment has its own identity and makes meaningful contributions to the production of scientific claims. Both machines and scientists are part of an extended feedback loop formed with the rest of society. Law explains the actor-network conceptualization of science as "a process of 'heterogeneous engineering' in which bits and pieces from the social, the technical, the conceptual and the textual are fitted together, and so converted (or 'translated') into a set of equally heterogeneous scientific products" (1992, 381). All components of this feedback loop are, according to Latour, actants, which have agency.

What does Latour mean by actors or actants having agency? In classical ANT, he emphasizes that actants impact one another symmetrically: "We do not know who are the agents who make up our world. We must begin with this uncertainty if we are to understand how, little by little, the agents defined one another, summoning other agents and attributing

to them intentions and strategies” (Latour, 1988, 35). From this quote, Latour appears to assume everything has agency and that all agents have equal role in defining the world. However, only humans observe, interpret and expose relations and interactions in their environment. Latour himself names scientists spokespersons of objects in the lab. However, he does not seem to recognize the deeper cultural implications of placing scientists in this position. As such, it is questionable whether we can ever understand how agents *define* one another, according to this version of ANT.

As his perspective on ANT changed, Latour (2005) endeavored to limit the potential misconceptions about the notions of actant and agency. In doing so he stressed ANT as a methodology, not a framework for understanding human-non-human interactions. In this vein, Latour (2005, 76) clarifies that ANT does not establish an absurd symmetry between humans and non-humans. Instead, he makes the case for *methodological* symmetry by emphasizing its role of including both humans and non-humans. Latour states that social sciences can thoroughly explain the world only if they take into consideration all entities that participate in an action. As such he insists that “*ANT is not the empty claim that objects do things ‘instead’ of human actors*” (Latour 2005, 72). In this way, Latour takes steps to make ANT more convincing to those committed to an anthropocentric or humanistic approach to agency. However, one lingering question regarding ANT rests on how all the entities subject to social scientists’ analysis must be considered actors in order to be given the credit they deserve for influencing social life.

Latour's attempt at redefining ANT seems to avoid settling on only one conceptualization of agency. For example, in *Pandora's Hope* Latour (1999, p. 190) claims that "agency resides in the blind spot in which society and matter exchange properties". At one point in *Reassembling the Social*, Latour defines actor something that "is made to act by many others" (Latour 2005, 46). Later in the same book (2005, 71), he brings forth the idea of an agent as something making a difference in the course of other agents' action, as such, acting *on* other agents in a *detectible* manner. This last perspective corresponds to the views Latour expounded in his earlier writing.

Taken altogether, Latour's views seem to favor a conceptualization of agency that conforms to the non-anthropocentric stance of agency (Knappett & Malafouris). In an effort to bring social scientists' attention to the power of materiality, Knappett and Malafouris (2008), argue, in line with Latour (1999), that agency lies in the interaction between humans and objects. They state that they aim to get away from the debate around who and what is an agent, by recognizing that it is situated in material culture. However, they themselves establish a baseline for how to conceptualize material agency by explaining that material agency is not the same as intentionality, as the notion of agency might imply. As such, the non-anthropocentric view of material agency only raises questions about the nature of agency.

One major critique of Latour's ideas of agency offers a different scope for the concept of agency. Kaptelinin and Nardi (2006) put a structure around Latour's attribution of agency to all humans and non-humans in the lab (Table 1). They define human agency as the "ability

and the need to act” (Kaptelinin & Nardi 2006, 241). For them the “need to act encompasses both biological needs and cultural needs” which only humans have (Kaptelinin & Nardi 2006, 242). Other organisms are limited to a biological need to act. Thus, they argue that non-humans (objects and organisms alike) can only have conditional agency and/or delegated agency. More specifically, some objects may have one of these two forms of agency if they, standing alone affect humans or non-humans in their surroundings. Kaptelinin and Nardi counter Latour’s notion of symmetry of agents by positing a hierarchical distribution, in which the position of various actors is necessarily asymmetrical. Although Kaptelinin and Nardi’s view of agency involves a change in scope of Latour’s notion of agency, they raise an essential concern, that perhaps even using agency from methodological considerations may stretch the utility of this concept. Their perspective represents a humanist determination of agency, as it recognizes the role of humans in acting out of cultural needs, their high standing in the hierarchy of agents, as well as, implicitly, their primacy in classifying agents. The typology is, nevertheless, quite cumbersome and may ignite debates around how to classify agents in the lab.

In contrast to the non-anthropocentric perspective on agency, which presupposes an outsider’s outlook on the lab and does not include the scientists’ view, classical anthropological work (Malinowski 2013), premises the humans as originators of meanings. This perspective is strongly linked to humans as originators of social relations and conceptualizations of sociality of objects that emphasize the role of material objects in social relations and the production of meaning (Fiske 1992; Law and Mol 1995; Knorr-Cetina

1997). Fiske (1992, 693), for example, notes that things derive their meanings from the social relationships enveloping them. In other words, “when we look at the social, we are also looking at the production of materiality. And when we look at materials, we are witnessing the production of the social” (Law & Mol 1995). An approach to agency that considers sociality, furthermore, accounts for cultural differences in humans’ interactions with objects, something the non-anthropocentric perspective fails to do.

Findings from the *Drosophila* lab suggest that assuming agency of the humans and non-humans in the lab prior to observing lab interactions not only de-emphasizes the lab as a cultural world, it may not be useful for understanding the relationships between humans, instruments and model organisms there. The notion of indeterminacy (Knorr-Cetina 1981) is powerful for conceptualizing the laboratory as a space of particularities, in which humans make decisions about their interactions with the objects around them. As such, they also make decisions about what is an agent and when. The concept of instrument-experimental object interface further supports this contention in relation to biological laboratories. The intersection between material objects and organisms may ignite reactions from the experimenter that are both specific to the experimental object used and interpreted through a social lens. This latter point brings to the fore various dimensions (e.g. affective, rational) of the relations between the scientists, their tools and the fruit flies in the *Drosophila* lab. In particular, it shows the ambivalence with which scientists in this case do, indeed, approach the question of agency, both their own and that of the objects around them.

Table 1. Kaptelinin and Nardi's typology of agency

Agencies	Agents	Things (natural)	Things (cultural)	Nonhuman living beings (natural)	Nonhuman living beings (cultural)	Human beings	Social entities
	Examples	Tsunamis, Northern lights, vernal pools, Martian rocks	Speed bumps, sewing machines, teapots, adzes	Grizzly bears, California poppies, truffles, protozoa	House cats, Dolly the sheep, GMO corn, Bourbon roses	Spinuzzi's traffic engineers, Miettinen's scientists, ANT's princes	World Trade Organization, ISO, Doctors without Borders, United Nations
Conditiona l agency	Produce effects	+	+	+	+	+	+
Need-based agency	Act according to own biological needs	-	-	+	+	+	-
	Act according to own cultural needs	-	-	-	-	+	+
Delegated agency	Realize intentions of (other) human beings	-	+	-	+	+	+

CHAPTER 3 RESEARCH METHODS

In order to document the interactions in the lab, I conducted participant observation with the fruit flies, the equipment and the scientists who use them for two months during the summer of 2013. This study included 25 people¹: four senior researchers (two female and two male), five postdocs (two female and three male), four graduate students (three female and one male), six technicians (all female), and six research assistants (four female and two male). The senior researchers are long standing employees of the laboratory in question. The post-docs receive their training with the expectation that they will establish their own laboratories in the future. The graduate students work mainly on individual projects for their doctoral dissertations. The technicians help maintain the genetic strains developed in the laboratory. The research assistants can have their own projects, but mainly help senior researchers, postdocs and PhD students with their projects.

According to H. Russell Bernard (1988: 148), participant observation involves “establishing rapport in a new community” and learning how to act in such a way that one easily blends in and behaves appropriately in that community. Although at times, due to my lack of expertise, I was unable to discuss their research at the same level as they discussed among themselves, I did manage to blend in with the researchers’ social community, especially as the coordinators of the lab allowed to work on an individual research project on *Drosophila*’s sensitivity to ethanol. My presence as a colleague in the lab helped the staff

¹ During my fieldwork, the lab also included two visiting researchers and two undergraduate assistants who came at about the same time I joined the lab. I excluded them from the study because they did not have much experience working in this lab. I also excluded all staff that did not perform tasks involving flies and equipment for manipulating them, such as the staff who made food for flies.

become more comfortable with me. After some initial skepticism, my presence became part of the routine and I lab staff would invite me often to observe experiments. From time to time, especially when somewhat sensitive discussions were taking place, some of the more humorous of the group would joke about the outrageous conclusions I might draw from their conversation.

I gathered qualitative information focusing on scientists' experiments, noting step-by-step the procedures they performed, the way they used the tools and objects around them, as well as how they manipulated the fruit flies. I informally interacted with the scientists, asking them to describe the actions they performed, noting how they talked about or to the equipment and fruit flies. I looked at the kinds of difficulties the scientists encountered noting unexpected events and the ways in which scientists reacted and talked about them. I also attended seven lab meetings to observe how scientists talked among each other about their experiences in the laboratory with the equipment.

Informal and semi-structured interviews provided more detailed accounts about their ideas concerning their lab work. The informal interviews took place during breaks, at lunchtime, after lab meetings, or during coincidental encounters, and they often included more than one interviewee. The semi-structured interviews took place during scheduled timeslots. I conducted 23 semi-structured interviews involving an interview guide (Bernard 1988) ensuring the discussion of the same broader topics with all interviewees in order to gather comparable data (see Appendix B).

During both types of interviews, I inquired, among other things, about the instruments scientists use most often, the tools they prefer to use, the concerns they have when using them, how they think about the usefulness of *Drosophila*, how they assess the validity and usefulness of statistical analyses, and overall their work in the laboratory. Re-phrasing questions in various settings and times allowed me to identify consistency or fluctuations in people's responses.

Upon gathering the data, I segmented the relevant portions of the interviews that contained information about scientists' concerns when using their tools. I used the topic of conversation (for interviews) or of descriptions (for fieldwork notes) as the unit of segmentation. I developed a coding scheme based on the patterns that emerged from perusing the data several times relative to my research questions: the kinds of instruments used, concerns regarding use of instruments and ideas about fruit flies. I later used the patterns to draw generalizations vis-à-vis the theory.

To validate my findings I conducted a lab meeting in which I presented my preliminary ethnographic account and received feedback from the laboratory staff and coordinators. I also presented my interpretations of their remarks, experiments and interview excerpts to individual staff members and I incorporated their feedback in my writing.

CHAPTER 4 THE DROSOPHILA LAB

4.1. Background

The study site included two adjacent *Drosophila* labs which work collaboratively. This means the labs share resources, and the scientists are allowed to use facilities within all rooms pertaining to the labs. Between the two labs, there are almost 30 employees. The number is not constant as research assistants and visiting researchers come and go constantly. The joint laboratory is now a top *Drosophila* lab in the US. One of the main characteristics of the lab that I noticed early in my fieldwork is researchers' propensity for helping each other. "We collaborate a lot. Other labs don't collaborate, but they don't excel" said a technician one day.

The *Drosophila* laboratory investigates the environmental and genetic factors that affect "quantitative traits" in *Drosophila melanogaster* (Welcome to the Mackay Lab 2013). "Quantitative traits" refers to physical characteristics or behaviors that are coded by multiple interacting genes. As such, the traits are complex. Examples of projects include, testing sensitivity to alcohol, measuring life-span, food intake, phototaxis and aggression. Given that grant sources usually prefer projects that may be applicable to humans, many of the projects emphasize the relevance of findings about the fly model for human benefits.

The two labs (Fig. 2) include four fly rooms where each researcher typically has his/her own bench (sometimes researchers share a bench, if there are too many people in the lab). A "bench" is basically a desk with a shelving hutch; there is one large drawer underneath the desk and several more on one side. Two molecular labs are adjacent to fly

rooms (see Fig. 2). They also have benches; however, the desks are taller (one has to stand or sit on a high chair when working at them). In two “behavioral rooms” researchers conduct most of their experiments with flies. Additional rooms contain freezers, refrigerators, incubators and an autoclave (a big, hot furnace for melting used vials, heating substances used in experiments or decontamination), a kitchen where the fly food is cooked and packaged, as well as miscellaneous offices.

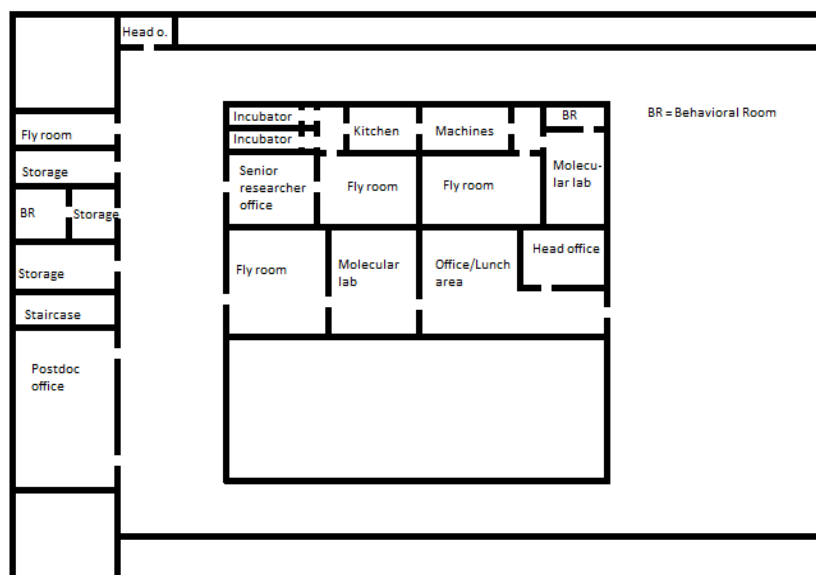


Fig. 1. Map of the lab (not drawn to scale)

Given their importance in experimentation, further description of the behavioral rooms is warranted here. The main behavioral room is small. Despite the confined space, this room often accommodates two experiments at a time. This description of the behavioral room creates an image of the lab as a space of improvisation:

There are two loud ventilators on the ceiling; four neon bulbs form a rectangle around the two ventilators. The room has controlled temperature and humidity. There are tables on three sides next to the walls covering their whole length. There are also two tall chairs in the room. On the table on the left, there is a white fabric box (lab staff call it a “studio”); inside, there is a vial-box turned upside down; on top there are three groups of three vials which are stuck together with tape; they’re turned upside down, disposed in a triangle to create stability for a red plastic plate on top of them. On each side of the box there are two lamps. At the corner of the table there is a box with a jvc camcorder on top of a paper bag placed in a vial box with several scattered empty vials. The other two tables also have empty vial boxes in the corners, but a large portion is clean for experiments. There are two nonfunctional lamps on each side of the door. Cotton plugs, old and broken rubber bands and other small laboratory paraphernalia are scattered on the floor, especially under the tables.¹

Research in the lab takes place in several stages. I focus on three which involve the systematic use of instruments. These stages are deeply interconnected and can blend into one

¹ Using ideas of agency there could be several interpretations of this space. In line with the ANT perspective, the limited space on the tables would cause scientists to arrange objects in a particular way. The tables would thus appear to have agency. However, it is the researchers who choose to store unused objects in the space available. Kaptelinin and Nardi (2007) would argue that lab staff delegate agency to the tables to hold objects. If the tables have agency, the unused objects on the tables should also have agency, as should the paraphernalia on the floor. Considering those as having some sort of delegated agency would then require a rigorous classification of objects with or without agency. This makes for a complicated theoretical framework. Kaptelinin and Nardi (2006) themselves have troubles classifying certain laboratory objects. This renders the utility of their fairly complex typology of agency questionable.

Another way to interpret the passage above is as follows. The description above shows that only the relevant space – the space on the tables used for experiments – is ordered. Parts of the rest of the space appear chaotic; however, this apparent chaos does not disturb the functionality of the space used. At the same time it may seem that it restricts the size of the space used for experiments. However, the researchers use the corners for storage precisely because they do not need it for experiments. Scientists make decisions depending on the laboratory environment (Knorr-Cetina 1981). In the behavioral room several experimenters distribute various objects in the space allocated, and at the same time conform to that use of space.

another. First, researchers conduct phenotyping or “behavioral assays” in which they test flies using particular sets of instruments, typically comprising vials (plastic tubes in which flies are kept), cotton plugs, and vial racks. “Assay” refers to testing or assessing the behavior of the object of study in particular experimental conditions. Each experimental design using flies has a protocol or a set of instructions for performing the assays.

The second stage is the molecular work, which involves extracting and sequencing DNA or RNA from the flies (see Appendix A). At this stage the scientists also follow protocols, step-by step instructions on how to manipulate the content of fly cells. The tools and machines typically used in this stage are pipettes, plastic tubes, buffers, 96-well plates, Polymerase Chain Reaction (PCR) machines, centrifuges and incubators. Buffers, substances that aid with particular molecular reactions, such as DNA or RNA extraction or the preparation for particular processes, such as PCR, are kept in big bottles, or in tubes. Samples, other small ingredients for mixes of substances and reagents used for PCR for instance, are kept in small tubes of 1.5 or 2 milliliters, or in the wells of plastic plates. The proper storage for these is in the freezer.

Whereas for behavioral assays scientists learn the protocols and subsequently try to perform them consistently, in molecular work, they must look at each step of the protocol every time they want to perform a particular process. Molecular protocols specify detailed points, such as quantities and temperatures that scientists must check carefully.

For molecular work, scientists may develop protocols for different molecular analyses in the laboratory, or they may acquire them with readymade kits. As far as readymade kits

are concerned, as one of the postdocs in the lab put it, “one size doesn’t fit all”. By this he meant that the readymade kits do not fit all the types of organisms. They are not customized for flies. As such the staff in the molecular lab must analyze each protocol and adapt it for fly DNA or RNA. Protocols from kits are typically very expensive, on the order of thousands of dollars. All the workers in the molecular lab emphasized the importance of high costs and how they manage to elude them when possible. For instance they would try to make their own buffers, rather than ordering kits for certain protocols, such as library preparation (see Appendix A).

The third stage I include in my reporting is data analysis. Researchers run the data from the behavioral assays from fly work and the DNA sequences obtained in the molecular work through a Genome-Wide Association Study (GWAS). GWAS helps make correlations between particular genes and traits observed during behavioral experiments. They use GWASpi (Pipeline). This procedure analyses all the information accumulated in the lab about *Drosophila* DNA sequences, RNA sequences, and proteins to yield a list of genes that may be relevant to the trait analyzed. Data analysis also includes descriptive and inferential statistics.

Some researchers include a fourth stage in their projects, which involves the confirmation of results. Confirmatory studies include going through the first three stages of a project again. In confirmatory analyses the phenotyping stage involves manipulations of the strains of flies used initially. At this stage the flies are often transgenic (see below). There are also confirmatory analyses using an outbred population of flies (see below). Researchers

conduct complex analyses comparing the results of the confirmatory study with results from the initial project.

At different stages of a project, as several people in the lab told me, there is a clear division of labor in the laboratory. Different scientists employ different equipment in the laboratory: equipment to monitor the behavior or morphological characteristics of *Drosophila*; equipment used in molecular work on the genes of fruit flies, and equipment used to model the traits monitored in experiments. Some scientists mainly conducted fly work and behavioral assays; some worked in the molecular lab, and some only did data analysis. There were several who alternated their work between the behavioral rooms and the molecular lab. Others, transgressed their regular routine, be it fly work or molecular work, only from time to time. PhD students, postdocs and senior researchers, and one technician all did basic statistics. However, these individuals generally did not do the complex phenotype-genotype correlations on a regular basis as only the GWAS specialists did.

My reporting draws very little on the preliminary stage to every project conducted in the lab. This stage is typically not readily visible. This is the process of developing the research question and the protocol or set of instructions for each behavioral assay. This process may begin with an interesting observation or with intriguing results from a previous study. Brainstorming sessions follow, involving discussions with other people in the lab, or with the head of the lab. Brainstorming leads to hypothesis generation. Further brainstorming leads to the experiment's design and invention of necessary tools. The researcher may conduct preliminary tests to ensure the experimental system is functional. These tests serve

as feedback for the workability of the hypothesis. The preliminary tests also inform the researcher about the suitability of the materials and tools s/he plans to use. Even after the preliminary tests, researchers actively cultivate feedback among the main research stages. Observations from one stage inform observations from the other stages, and researchers can go back and forth between their behavioral assays and results from statistical analyses. In this way, the three main stages are part of ongoing fluid processes.

The last step of a research project, is the write up. Research is bound to be communicated, either to build on basic science or to contribute to knowledge that can be applied to human health. For *Drosophila* research, many experimental designs address the quest for basic knowledge about genetics, and *Drosophila* genetics in particular. At the end of a research project, the researchers also write and submit to the funding institution a project report.

4.2. *Fly Stocks*

Before discussing the nature of the interaction between scientists and flies, it is necessary to introduce the types of flies used in the lab and the kinds of fly work scientists conduct. Fly work consists of several types of tasks: the construction of experimental populations (i.e. creating inbred strains through controlled mating and outbred populations of flies through random mating); culture and maintenance (which involves transferring or “tipping” flies to new vials or bottles, feeding outbred populations) and analysis (which entails sorting and collecting flies for various experiments and conducting behavioral assays, or phenotyping).

Researchers carry out these tasks as they work with different fly types. Most fly stocks are inbred or outbred lines. Some stocks are wild-type. Wild-type refers to flies collected from the outside environment, which have not developed mutations in the laboratory, only manifest the ones that also occur in the wild (such as white eyes or curly wings). Stocks with recognizable mutations have been reared in *Drosophila* labs in some cases for decades. The lab also has transgenic flies created through different genetic engineering techniques.

Inbreeding is a process of creating nearly homozygous individuals (i.e. having the same version of a gene on both chromosomes in a pair for almost all genes). The main inbred lines are 192 lines entitled *Drosophila* Genetic Reference Panel (DGRP). These are stocks of flies derived from wild populations of *Drosophila melanogaster* gathered from the Farmers' Market in Raleigh, NC. It took 20 generations of inbreeding flies (mating flies which are brothers and sisters) to create the DGRP. The lab keeps or creates other inbred lines, using them for different purposes. Rose lines, for example, are inbred lines used mainly for mating and senescence assays. *Drosophila simulans* is a different species of *Drosophila* that was inbred during my time in the lab and was designed for comparison studies with *melanogaster*.

Creating inbred populations has caveats. One day, I observed one of the senior researchers at work while he talked about the development of DGRP: "Of the 900-1000 lines we started, 345 made it through inbreeding." This is a demonstration of the power of lethality and sterility. Even if lines made it through inbreeding, they can still perish if they are not

healthy enough. For instance one of the postdocs described how flies from one line have abdominal problems, which is thought to be the cause of their short life. Those flies breed badly as well.

When creating inbred lines, natural mutations can be useful as a marker for a particular line. Some of the older members of the lab enthusiastically recognize various mutations and appreciate them. One day one of the senior researchers was selecting pairs of flies that would produce the inbred lines of *Drosophila simulans*.

Suddenly I hear him rejoicing at something: “Woowee! Alright!” He smiles and is obviously happy about something. “Is that a new mutation?” I ask. It was. “Vermillion eye; I’ll make a note of that on the vial”, he says proudly.

Outbreeding involves mixing various inbred strains and letting them interbreed freely to create a heterozygous population. *Flyland* is a population outbred from DGRP and used for confirming experiments on DGRP lines. Furthermore, numerous studies create ad-hoc crosses – for instance between inbred lines and control lines, or inbred lines and transgenic lines – relevant for particular research questions.

Transgenic flies, or “mutants”, as people in the lab often call them, are another essential type of fly. They can be used for regular assays or for confirmation studies. A transgenic fly can be produced in several ways: gene silencing, gene knockout, gene overexpression or allelic substitution. Gene silencing involves inserting in flies a synthetic gene that interrupts the expression of a gene of interest (a DNA sequence coding for a particular trait or behavior). The outcome of this process is that a particular gene does not

carry out its usual protein production. As such, by silencing a gene, a phenotype will be altered or no longer present. Constructs that can help with gene silencing are RNAi and the GAL4/UAS system. Gene knockout is done by inserting *p* or Minos transposable elements that physically interrupt a DNA sequence and insert themselves in the gap (see Appendix A). Researchers also use the GAL4/UAS system to cause gene overexpression. Gene overexpression refers to stimulating a gene to become expressed more often than normal. Another common way to create mutants is through allelic substitution. Allelic substitution involves replacing one version of a gene with another variant for which the phenotype is known. This changes the fly's phenotype. According to a technician in the molecular lab, this "is super-super functional testing. [...] If you do this, nobody can question you". This testing helps confirm GWAS results and thus increases the reliability of a study.

The lab orders some transgenic lines directly from other laboratories (Harvard, Vienna, Bloomington, Kyoto). Sometimes, the lab creates particular constructs, especially for allelic substitution, in order to suit certain experiments. They deliver the constructs to Duke University, to a facility for germline integration, where the constructs are inserted into fruit fly embryos which develop into transgenic flies.

Much fly work takes place at the benches in the fly rooms. On the shelves, the lab staff have some of the stocks of flies they use. Researchers also keep big stocks that require a specific rearing temperature in incubators. Given the number of vials and bottles with which they deal, labeling vials is essential. Sharpie's (markers) of different colors and thickness, are

present in every scientist's drawer. Scientists label all vials with the name of the line of flies, the date when they put flies in the vial, and where appropriate, the temperature.

CHAPTER 5 AMBIVALENCE AND FLY WORK

The above description of the laboratory and of fly stocks establishes the context for the scientists' work with fruit flies. *Drosophila*'s transformation begins in the fly rooms or in the behavioral rooms, where scientists use particular instruments to test *Drosophila*'s behaviors or measure certain traits. Below, I elaborate on researchers' concerns with respect to their interaction with fruit flies and the equipment for fly assays. Their concerns illuminate the importance of their perception of the agency of objects and flies. I detail this idea by presenting different dimensions of fly work, including the role of scientists' choice and invention of tools in affirming their own agency. The particularities of the fly influence scientists' perception of the insect as an agent in the lab. The interaction between fruit flies and instruments which informs experimental design also impacts how scientists confer agency to the flies rather than to the objects with which the flies interact.

In preparation for behavioral assays, scientists sort flies for experiments, since typically a project involves separate replications for the two sexes. Sorting and collection always takes place in the fly rooms. To sort flies, researchers use carbon dioxide (CO₂) to anesthetize the insects. Carbon dioxide flows through plastic tubes from large storage tanks to each bench. At the benches the gas flows through a tap into small plastic boxes with porous surfaces called "fly pads" or "CO₂ pads". Flies placed on the pads become anesthetized within several seconds and remain anesthetized for the duration of the sorting. The work needs to be done quickly as flies that under the influence of CO₂ for more than a few minutes may become sterile or die. These issues become important depending on the

experiment in question. For instance, sterile flies would be useless if the researchers need to carry out a cross. Two members of the lab systematically use ether as an alternative for anesthetizing the flies. Scientists pass flies through a funnel containing ether and then place them on a metal plate or ceramic tile. They must sort flies before they wake up and fly away. For some experiments flies' exposure to CO₂ or ether may be detrimental. In this case, scientists place a container with flies on dry ice, and sort flies while they are in a "chill coma".

Depending on their nature and requirements, experiments can either take place in the behavioral room or in the fly room. For instance, researchers can store flies in small bottles with special solutions, and after some time, remove their cuticles with the help of special instrumentation in the fly room. They may also anesthetize flies and put them under the camera of a special dissection scope and photograph their thorax in order to later measure it.

In the behavioral room, the researcher can, for instance, put flies to sleep using a solution called Fly-nap (a product designed not to interfere with flies' heart rate, as does CO₂ or ether). The researcher then sticks the insect's wings to a small polystyrene plate and counts its number of heart beats per minute. In other assays taking place in the behavioral rooms, researchers may put flies in containers, such as vials, and observe or film their mating or aggressive behaviors.

Recording fly behavior or traits is one dimension of *Drosophila*'s transformation. Usually the researchers conduct experiments on a series of fly strains and record the results on printed tables or in a lab-book. They each design their own tables depending on the nature

of the experiments, and the information they need to record (I even saw customized sheets for males printed in blue and females in pink).

5.1. *It's not the Tool's Fault*

I describe below the instrumentation for fly work and scientists' creativity in the fly room. My observations reveal the particular nature of interactions between scientists and instrumentation in the *Drosophila* lab studied. On the one hand, scientists' practice and most of their speech suggests they take responsibility for their actions in the lab by choosing, manipulating instruments and creating new ones. As such, they affirm their own agency. On the other, their speech about the instruments also reveals a tendency to anthropomorphize objects, as such, conferring agency to them.

A closer look at some crucial instruments for fly work and the ways in which scientists talk about and use them illustrates the ambivalence towards the agency of objects. People in the lab sometimes develop their own instruments to carry out fly work, taking objects from around the lab and elsewhere and putting them to new use. Researchers often expressed their preference for working with certain instruments. Importantly, they typically took the needs and characteristics of the fly as a point of reference when developing new instruments. They also sought to make something that is convenient for them and that suits the purposes of their various tasks. One day, one of the senior researchers told me: "Our general rule is you try to find the conditions that work best for you¹, and you keep doing the trick again and again". This is especially the case with using fly sorting instruments. Many

¹ And importantly, for the flies as well, as mentioned above, and as the next subsection shows. The flies are agents in practice; consequently, researchers must consider their behavioral peculiarities.

lab workers used paint brushes, which come in different shapes and sizes according to preference.

Senior researcher: *“Ooh, you have to have a good brush, to sort them, right? Some people use metal spatulas, [...] but I still prefer brush. Brush would be like... [...] firm and long, bristles; should be tight. So natural bristle is better than artificial one.”*

The senior researcher describes the characteristics of the type of brush with which she prefers working. Others in the lab may have different sets of preferable characteristics. Some researchers emphasized their special relationship with their favorite brush which they trimmed themselves, and have used, in some cases, for many years. This emphasis on the relationship between scientist and objects corresponds to a local manner of producing sociality, as scientists give special meaning to their tools (Law & Mol 1995).

Other people in the lab preferred small metal spatulas usually found in the molecular lab and used for scooping out powders needed for solutions. Several of them used sculpting tools that one of the technicians bought from an arts and crafts shop. The sculpting tool has two ends, one flat and straight and one bent and narrow. One technician discovered that a dental instrument may suit her purposes. The instrument is a small metal rod with a small hook at the end that dentists use to detect cavities. According to her, it is easily sanitized and “It’s so precise, it’s so fine... It works like a charm”. By these remarks the technician was not affording agency to her dental instrument. On the contrary, she was emphasizing the physical qualities that are convenient for her purposes.

As I observed one of the postdocs collecting a set of flies for RNA extractions, we discussed the idea of having a favorite tool. He explained that depending on the project on which he works, he may use the aspirator, the sculpting tool, the bent brush that he made, or a short brush. The aspirator is a long pipette attached to a transparent plastic tube with a small hard plastic mouthpiece at the end. At the intersection of the tube and the pipette, a small screen is inserted to stop the flies from going through the tube in the experimenter's mouth. By breathing in through the mouthpiece, one can pick up runaway flies. One of the scientists brought the aspirator years ago from Japan, his native country. Throughout the years, many others in the lab adopted it.

The postdoc also uses the sculpting tool for work that involves avoiding the contamination of the flies collected. When sorting flies for RNA or DNA extraction, the scientists must be careful not to let the flies they are collecting touch objects that can contain the remains (and therefore, the RNA or DNA) of flies from other lines. Thus, all the objects with which the flies come in contact must be sterile. The postdoc also uses trimmed brush and a bent brush (to gently scoop out flies that are stuck in the food). He made the latter by keeping a brush in a vial with ethanol for several weeks.

Bob starts telling me about the advantages of the sculpting tool. "It's easy to sterilize... Metal is useful. You can put a flame on it" (He raises the tool in the air with his left hand and with the right hand moves an imaginary flame under it) [...] Bob continues to reflect on the idea of favorite tool: "Favorites are hard to qualify. Each one has its purpose". He says he's clumsy at scooping. Normally he uses the aspirator a lot. However, it is not useful now,

because it's not sterile, remains from other flies could be inside and can contaminate the sample. "For DNA work I use this guy" he says showing me the sculpting tool. For normal tipping he uses the bent brush, and for sorting, the small brush [...]. He opens the drawer, which is full of various office and small lab tools: "I'm definitely a tool guy. Each tool has its purpose. I'm a functionalist. What works best for a particular job, that's the tool for me".

The postdoc's use of "this guy" to refer to instruments at his bench, might suggest he is anthropomorphizing the instruments. Although the postdoc probably does not think of his instruments as having ability to act, his language, obviously playful, would qualify them as collaborators. By emphasizing his personal characteristics, such as clumsiness, and his power in deciding the purpose of each tool he is affirming his own agency. He commented later: "Obviously the agency is in the scientist not the tools, but a scientist without tools can't do the work any more than tools without a scientist". With this statement the postdoc does not imply some symmetry between him and his tools. He simply indicates his sociality with his tools, in that he needs his tools to be in his social world.

Finally, the dissection scope¹ is another critical tool, used for separating males from females. However, not all people in the lab use it. One of the senior researchers usually does not use the dissection scope as she is confident in her ability to distinguish between sexes based on many years of experience. Other scientists use the dissection scope all the time as they have become used to maneuvering the flies under the lenses. Yet other people in the lab

¹ In the lab there are several types of scopes. Some assays require more advanced dissection scopes, or require accessories for the basic scopes, such as goose-neck lamps (small lamps that have a long flexible neck). There is a dissection scope with a camera used for photographing flies for different projects (thorax measurement, abdomen pigmentation, etc). There is also a microscope with video camera that is connected to a computer, which facilitates the observation of flies up close.

resort to the dissection scope only when it is very difficult for them to distinguish between male and female flies.

Choice about the use of instruments reinforces the notion that humans are the agents in their network with the objects in the lab, as the use of objects depends on their skill, habituation and assessment of the circumstances of their tasks. Often, preference for certain objects is manifested depending on how well a scientist is able to handle an object. For instance, some people in the lab would not use a brush, because flies get caught in the bristles of the brush. Others know how to avoid catching flies in the bristles, so they always use a brush. Preference is also related to habituation. Some people have used a tool for a long time so they continue using it, as is the case of the experimenters who use the dissection scope all the time. A third mode of preference, as the example of the postdoc shows, relates to function, which leads to using a variety of tools irrespective of whether it is convenient for researchers to manipulate them. For each type of preference one may argue relative to agency that the object as agent imposes a particular behavior on the human agent. However, the object does not willingly impose a restriction on human behavior; nor does the human delegate the object to impose a restriction. It is only humans' idiosyncrasies, and agency implicitly, that call for the use of particular objects in distinct ways. It is nevertheless true that the availability only of certain instruments restricts researchers' choice.

Although not part of the first stage in the transformation of *Drosophila*, the invention of instruments is relevant to it and is connected to how the scientists in the lab (especially PhD students and postdocs) emphasize their agency in relation to their instruments. One of

the postdocs, a very creative and playful personality, developed a new version of an instrument for testing aggression in flies. It is a rectangular, transparent, plastic box with 24 circular wells which accommodates two male flies that engage in aggressive behavior. She called it the “flydiator arena”. She worked on it for about half a year, constantly refining the instrument, and consequently improving her assay.

“So I was just kinda looking around the lab and I saw these tissue culture plates and I was like, ‘Well that seems way more efficient than looking at the vials’. And so I started the assay by just figuring out how to get flies into each well of a 24 well tissue culture plate. And the hardest part of this was figuring out how to knock the flies out and then how to film them, ‘cause the video camera that we had wasn’t good enough.”

Notice the way the postdoc imposed a value judgment on the tools she had been using, as opposed to the ones she could use instead: vials were not as efficient as the wells seemed to be. Importantly, her work involved trial and error. The first time she described to me the development of the arena she concluded: “I’m like an engineer”. Discussing with another postdoc the importance of creativity in the lab, he stated: “Creativity is something that a scientist must have” [...] Without creativity you’ll do good work, but you won’t do great work [...] Sometimes you’ll have to make do with what you have”. The way these two postdocs think about their creative work in the lab is, ironically, in line with Lévi-Strauss’s (1966) description of the *bricoleur* (as opposed to the engineer), someone who uses the objects and tools at his or her disposal to build something functional. The quotes above reinforce the idea of the scientist as primary agent in the lab.

Objects, by their physical properties (such as texture, size, smell, chemical composition etc) or by general physical phenomena (gravity, inertia, electrical charge etc.), can resist human manipulation. Scientists are aware that their desire to use equipment in a particular way does not always correspond to these qualities. The remark of one of the senior researchers is relevant in this sense: “I usually get frustrated if things don’t go the way I want them to, but it’s not the tools’ fault. It’s my fault. So I have to reshape the tool”. The tool in this interaction is obviously a non-agent. The researcher thinks of the tool as an entity that should be entirely under his control.

The availability of particular instruments for fly work in the laboratory brings to attention the local character (Knorr-Cetina 1981) of the practice of the scientific community observed, which can frame the discussion of situated agency. Scientists use lab resources or external resources that they seek independently in order to develop the most convenient conditions for their fly work. Choice, as shown above, indicates researchers’ affirmation of their agency, and implicitly, the treatment of instruments as non-agents. In their speech, however, scientists have ways to anthropomorphize objects, thus attributing agency to them. Through scientists’ creativity and improvisation with materials alien to the lab, such as the sculpting tool, the fly room becomes a customized environment. Furthermore, the building of new instruments combining lab resources suggests scientists’ critical engagement with the objects in the lab. Again, by stressing the intellectual effort they put into their inventions, researchers emphasize their own agency.

5.2. *The Fly Tells You*

The interaction with flies is different than the interaction and perception of tools, both at the practice and speech level. I now turn to analyzing the perception of flies, and scientists' concerns with respect to the behavior and their handling of flies. Scientists think of *Drosophila* as a tool, a non-agent that they can manipulate. However, in practice, and when discussing the aspects about which they are mindful when working with flies, they emphasize the agency of flies.

First, as model organisms, flies are conceptualized as non-agents. One day, after discussing with one of the senior researchers the strategy for creating a new genetic construct for a transgenic set of flies, one of the heads of the lab said with content: "*Drosophila* - we have such nice little tools!". Further, one of the senior researchers told me one day: "They are good study organisms: there are many stocks available; they are easy to manipulate genetically; it is easy to produce transgenics. Also, they share metabolic pathways with mammals and other vertebrates". This description suggests the fruit flies' perception as an object that scientists can change at their discretion. It appears that as tools they are devoid of agency.

Although they all perceive flies as tools, the lab staff do have affectionate terms for them, usually marked by humor: "little buggers", "crazy flies", "my children". One day as I was passing through a fly room I spotted one of the postdocs looking at a vial and exclaiming. "You flies are silly" in a playful tone, like talking to a child. Another time, when working at my bench one of the assistants spotted a fly with different eye color in a vial full

of DGRP lines, which always have red eye color. He showed the vial to his supervisor who remarked in an amused tone: “Stupid flies”. These names and ways to address flies directly highlight the attribute of flies as agents: as living organisms with which scientists interact in a distinct way than they do with objects and instruments in the lab. The fact that researchers address the flies directly does not primarily suggest that flies are agents. However, the frequency and the marked affective dimension involved in this interaction that suggests flies have a different status than material objects in the lab; the researchers acknowledge their special status.

There are three main concerns related to the manipulation of flies that further suggests their treatment as agents: timing, cross-contamination, and actual handling. Timing is one of the most crucial concerns in fly work. When working with flies, lab staff depend on the “fly time”. There are two ways in which fly time is important. First, the scheduling of a project depends on the time when flies from the needed stocks emerge. Sometimes, flies’ hatching can be planned ahead. However, when doing crossings between strains, experimenters may not get enough flies for a project, if the crosses are not successful. The second way in which researchers depend on fly-time is related to flies’ circadian rhythm. Flies behave differently throughout the day, so experimenters should perform assays consistently at the same time throughout the duration of the research in order to limit confounds¹.

¹ More on experimental design in the following subsection.

Scientists are also concerned with cross-contamination¹. The most important is contamination of a strain of flies with genes from another strain. In the lab there are stray flies which can land in uncovered vials and lay their eggs in the food. If flies from another strain are transferred to the vial, and they mate with the flies that hatch from those eggs, the result will be hybrid offspring which are useless for experiments. This is why researchers must be extra careful about keeping empty vials covered at all times. Some prefer cheesecloth, which can cover all the empty vials in a box. Others are very particular about covering empty vials, so they use cotton plugs for each empty vial. One inventive technician devised a method to keep stray flies off her fly pad.

Rosy shows me her “flyshield 4000” which is a transparent rectangular lid, about the size of her CO2 pad (about 9x12 cm). She says it’s the top of a 96-well plate that was a throw-away. On the lid she wrote with a black marker the name of the instrument and made four drawings of circles with flies crossed by an oblique line. That is to suggest that flies are not allowed; a sort of a stop sign for flies. At the bottom, under the name of her invention stands written: “Keep yo’ stray assess off!” She says this is her Southern touch to her flyshield. Rosy uses the flyshield whenever somebody comes in and wants to talk to her, and she has to turn away from sorting flies on the CO2 pad. She covers the flies so that no fly that is freely moving around would accidentally fall on the CO2 and contaminate her “treasured babies”. “Cos

¹ The lab staff are also concerned with two other types of contamination. One type is with mites, small parasites which are attracted to decomposing matter, like food that is degrading. For this reason, scientists need to make sure they are constantly transferring, or in lab language “tipping”, their stocks of flies to new vials with fresh food. After several days of having been kept in the same vial, flies and particularly the larvae from the eggs they lay dig through the food softening it. Combined with fly excrements, and decomposing fly bodies, the medium becomes attractive to mites. The other type of contamination is with mold; high humidity can facilitate the development of mold inside vials or bottles.

you don't know if something lands on there and voilà!" She raises her tone in a declamatory manner expressing the imminence of a fly landing on the CO₂ and contaminating the line. Now she says she is working on a prototype for flyshield 5000.

The story of Rosy's flyshield offers a surprising type of interaction between scientists and flies: not only do they sometimes talk to their flies; they can even place signs for them to follow in the lab! The humor in this relation between scientist, flies and tools illuminates a ludic dimension of sociality in the lab; the flies' agency is acknowledged through their pretended treatment as people who can read. Further, the contingency of lab interactions is illustrated through the technician's reference to the uncertainty of flies flying on her CO₂ pad.

Handling flies, although it may seem easy, is a skill one has to work on in order to perform experiments well. For proper handling, it is instrumental to know fly anatomy and typical features of fly behavior. Although experimenters have different styles of tipping flies in terms of how they maneuver the vials, they seem to have one thing in common, which I realized after complaining to some of the senior researchers that I always managed to lose flies. "If you handle gently, the flies will not be disturbed" one of them said. She meant that by tapping the vial fast enough and with precise wrist movements, the flies, even though obviously disturbed by the shaking, will not have time to fly up as they have the tendency to do. The researcher then showed me how she gently tapped the vial on the soft pad while connecting it with another vial. The flies in the vial that she was tapping were hovering

above their food, close to the bottom of the vial. After several seconds, after the flies from the other vial fell inside, she quickly covered the vial with a cotton plug.

At one point I asked one of the senior researchers how much CO₂ I should give the flies. He responded: “Ask the fly! [...] The fly tells you; just look at the fly”. He meant that if the flies continue to move, then the flow of CO₂ must be increased. When I observed the same senior researcher’s assay on the mating behavior of flies across species, he told me he put males in the experimental vials first, because they are “more tricky; more active”. As such, it is easier to transfer males before the females. Tipping males after the females would most likely result in males escaping.

One PhD candidate was mindful about fruit flies’ physiological needs when she performed the Capillary Feeding Assay (CAFE) for measuring flies’ food intake . Before eating flies go through a starvation period. One day I watched her perform and explain her work.

She pours the agar (a transparent liquid that becomes jelly after cooling down) from a flask into vials. It’s a seaweed based protein that has no caloric value. She explains her reasoning behind using agar for the starvation period of flies: “If you’re a little insect, acquiring water is one of the biggest challenges.” Lacking water leads to desiccation. Maggie emphasizes that offering water to the flies during their starvation period is a critical point in her experiment, and a major addition to the original setup for the CAFE assay that other researchers presented in a paper in 2007.

The way the postdoc talks about the challenge insects face in relation to acquiring water underlines empathy with the insect by using the pronoun ‘you’ and the diminutive “little insect”. She thus affords agency to the fly. Her speech marks the production of sociality with the model organisms in the lab through the emphasis on affect.

As shown above, qualifying flies as tools indicates that scientists think of flies as non-agents, entities they can easily manipulate. However, the distinct ways in which researchers talk about and handle the flies as opposed to the ways in which they talk about material tools suggests their treatment of *Drosophila* as an agent. Flies’ characteristics are more complex than the characteristics of material tools and affect in a large measure scientists’ behavior and the set-up of experiments (as do timing, flies’ tendency to fly upwards and their need of water). Furthermore, scientists refer to fruit flies using affective terms, which suggests a different hierarchical position of *Drosophila* in the lab compared to material tools. Further, communicating with flies, in oral and written form, suggests a powerful manifestation of sociality with the flies.

Importantly, the relation between humans and flies most often includes instruments, and as such, an epistemic dimension. Scientists respond to what the fruit flies tell them: the flies’ reactions to the material tools with which experimenters manipulate them (e.g. CO₂). In their interaction with instruments flies have their own agenda, namely to fly away. What “they tell” the researcher is therefore not what they need, but rather the intensity of the effect of the instrument. They give an indication of how much more intense or less intense the

effect must be. This instrument-experimental object interface (Rheinberger 2010) is the focus of the next subsection.

5.3. *These Flies Are so Temperamental*

In addition to scientists' concerns related to flies, concerns regarding experimental design and implicitly the interaction between flies and the instruments used to manipulate them are important. Experimental design refers to concerns for sound data collection, establishing the sample size for the population of flies studied, the treatment to which the experimenters will subject the flies and the number of controls and replications. It also includes consideration of managing potentially undesirable factors that can affect the experiments (confounds). I illustrate how scientists afford agency to flies in the context of the experimental system and give instruments the status of non-agents.

Researchers design all phenotyping assays in consideration of later statistical analyses. For this, scientists do several replications for each strain, inbred, outbred, or transgenic, for reliability purposes. Projects may have different units of replications. In some projects the individual is the unit of replication; in others, the unit can be a group of flies. Furthermore, experimenters test males and females separately, because typically there are sex differences in physical traits and behavior. The number of replications is decided partly based on how much variability the trait has. Some researchers use formulas to determine the number of replications. Others use protocols that previous researchers used.

Experiments must be high throughput. This refers to conducting a high number of replications on all lines available in order to increase the statistical power of tests. Each

experiment must have a control group. Control¹ refers to a separate set of replications on a strain of flies that acts as a standard of reference. For particular assays, especially ones that use mutants, increasing the number of controls, results in increased statistical power.

A major concern with respect to experimental design is consistency and limiting confounds from the data collection process. The scientists are very mindful about potential environmental factors that can interfere with the behavior of flies during an assay. This concern spills over into the organization of and scientists' behavior within the experimental system. Experimenters always keep in mind that they have to be consistent when they perform their experiments. One day I was watching videos of the "Aggression Assay" with one of the PhD students. He conducted the "Aggression Assay" using a version of the "flydiator arena". He calls the arenas, plates.

"We try to account for environmental variation so that hopefully what we get is gene variation" Jim explains. He shows me a plate on which the flies in the middle are fighting, while the flies on the sides are eating and are not active. Jim explains that the food around the margins of the plate in which flies stayed during the night could have dried out. When flies were introduced to fresh food right before the assay, they focused on eating instead of fighting. "These are moving around and these are not moving; so this [...] is a problem. I may not even use this". Another problem Jim points out is the position of the plate with respect to the experimenter. "Flies that are closer to the observer can be more affected by his movement; that's why we put them in the white box" (he refers to the studio on one of the

¹ There are particular fly strains created in the lab that have a long-standing role as controls, such as *Canton-S B*.

tables in the main behavioral room). “So there’s a million different things that can affect behavior”. Now we observe a plate with all the flies on it in the right position and displaying the expected aggressive behavior. “We may have a great assay” Jim says.

The notes above show the importance of the interaction between flies and the flydiator arenas, including the multiple scenarios that can emerge from conducting this assay. This interaction is what Rheinberger (2010) pinpoints as a critical source of insight for the researcher, off of which he or she can make epistemic decisions about the assay conducted. In the context of the excerpt above, the experimenter demonstrates keen critical thinking about the interactions between flies and the plates; he also manifests his power of decision-making concerning which plates to choose.

For the “Aggression Assay” of the inventive postdoc from Section 5.1., the negative experience with a previous similar project determined her to develop the new experimental design. From that assay she realized that she needed to reduce the environmental variance. To do so, she decided to “test all the flies in as similar conditions as possible – same batch of food, roughly the same number of generations in captivity, same amount of time in 20 degrees, filmed around the same time of day, etc.”. The “flydiator arenas” allowed for all these requirements, as well as an increase in the throughput, which gave more power to the statistics. This organization of the assay meant that the tests on all the DGRP lines would take only ten very busy days instead of 100, as it took in the original protocol for the assay.

Importantly, in spite of these concerns and the measures they take to increase the reliability and validity of their assays, researchers acknowledge that many factors influence

statistical analyses. The way in which the aforementioned postdoc admits this aspect also gives an indication of her position with respect to flies' agency:

“But also these flies... they are soo temperamental! So they behave differently if the humidity is different; they behave differently if the temperature is different. There are probably all kinds of weird seasonal factors that we think don't matter because they're in a building, but might matter; we just don't know.”

Scientists are, as such, conscious that despite having accumulated a tremendous amount of knowledge from this insect, *Drosophila*'s behavioral quirks surpass researchers' ability to completely overpower it, and comply entirely with the requirements for consistency.

Considerations for experimental design further demonstrate the role of the fruit fly as an agent whose needs and complex behaviors the researcher must understand in order to construct a reliable assay. There are, however, certain behaviors that are not even well researched and confirmed, which may pose problems. As shown above, the researchers transform objects or learn something about objects only through the flies' interaction with them. The example of the flydiator arena illustrates that researchers adapt their work according to the characteristics of the intersection between instruments and fruit flies (Rheinberger 2007, Latour 1999). The intersection imposes restrictions on the way researchers should manipulate objects. However ultimately, the scientist gives meaning to the intersection and understands it in a way that may be different across lab, or among different scientists.

CHAPTER 6 AMBIVALENCE IN THE MOLECULAR LAB

In fly work, flies and instruments are sometimes agents, sometimes non-agents from researchers' perspective. Molecular work is another critical stage in the transformation of *Drosophila*. As mentioned earlier, it is not necessarily consecutive to fly work. Scientists break the fruit fly into its smallest components and extract their genes. As in fly work, scientists display ambivalent notions of agency relative to instrumentation. Sometimes, their speech suggests they perceive instruments as agents. However, instruments are non-agents while scientists manipulate them. Interestingly, scientists appear to treat fly genes, both in speech and practice, as non-agents. Scientists' speech affirms their own agency through an emphasis on the responsibility they have at each stage of a protocol.

Scientists store the flies they need for molecular work in a freezer. Subsequently, they crush the flies in a mortar with a pestle or in tubes with magnetic beads. In this way, flies become DNA or RNA samples through a series of extraction procedures that require mixing fly components with buffers and reagents and filtering the substances obtained. DNA (deoxyribonucleic acid) is a macromolecule in the shape of a double stranded spiral, which encodes genetic information used in the development and functioning of organisms. RNA (ribonucleic acid) is the complementary replica of one strand of DNA; it exits the cell nucleus where DNA is located and enters the cell cytoplasm where different organelles convert its information into proteins.

Numerous processes that take place in the molecular lab after DNA and RNA extractions: genotyping; library preparation; gene cloning; Polymerase Chain Reaction

(PCR); microarrays (see Appendix A). Sequencing is another crucial procedure for the lab in general. It involves finding the sequence of nucleotides that compose a gene. This lab outsources sequencing to a specialized service. For the standard inbred and outbred lines, flies' DNA and RNA have already been sequenced. In this lab, genotyping is one of the most crucial procedures. It identifies and compares SNPs (single nucleotide polymorphisms) in different fly lines. SNPs are the basis of genetic variation between organisms in the same species representing the series of nucleotides different between alleles (different versions of the same gene). The data are stored in a database and the GWAS Pipeline retrieves them any time researchers need new phenotype-genotype correlations for a project.

6.1. You just Need to Follow the Recipe

To ensure the success of a process, researchers must, most importantly, follow the protocol. It involves adding the substances needed at the right time, following instructions for the incubation of substances, keeping the right temperatures, setting the right number of rotations in the centrifuge etc. One of the technicians, very enthusiastic about molecular work, told me that “molecular work is like cooking, you just need to follow the recipe”.

Furthermore, researchers are mindful about consistency and measuring quantities correctly. For instance, errors accumulate due to the quality of the tips, as well as the pressure from pipetting multiple times. Consistency is especially a sensitive matter when working with multichannel pipettes, which have multiple tips, attached horizontally to one pipette. One always must ensure that each tip takes the same amount of substance. Often

researchers lift the instrument a little above their head close to the level of the eyes and carefully observe whether the quantity is similar among tips.

Scientists also emphasized timing as a critical aspect of molecular work. Protocols involve a strict schedule for the performance of each step in a molecular project. One needs to pay attention and prepare for the following steps. A technician told me: “You have to plan out the tasks that you’re doing at any given time if you’re dealing with molecular tasks, like anticipate what you’ll be doing in the next couple of steps on the line. And do time management.”

People in the molecular lab are also careful about cleanliness and avoiding cross-contamination. The particles with which the molecular scientists are working are microscopic. Researchers may not discover errors that lead to a mixture of one sample with another sample until the end of a process. Consequently, keeping all utensils sterile is mandatory:

Senior researcher: *“Cleanliness – number one. Especially, when working with RNA, [...] you need to make sure that the laboratory bench has been treated with the special spray called RNAzap. This spray will destroy RNase, thus preventing the degradation of RNA that you’re working with; always wear gloves.”*

In the example above, the researcher describes the threat of RNase, an enzyme that degrades RNA. When working with RNA, scientists take precautionary measures to prevent the degradation of the RNA sample. By talking about the degradation of RNA, the researcher

seems to refer to RNA not as something that has agency, but rather as a non-agent, that degrades, like a substance.

Lastly, researchers are mindful about the actual handling and positioning of the instruments to ensure that there are no mistakes in the placement or combination of substances. The focus in the molecular lab is on manipulating the DNA or RNA according to the protocol, and not making mistakes.

“I usually have my fingers like outstretched, and you add solutions, you count one, two, three, four, five, six seven, eight, until you’re done. Just to be sure that you pipette solutions that you need, you don’t think about anything else. If you have a gap, you usually read the protocol, what to do next. You don’t care about the flies or anything at that point; you maybe care whether you have enough solution left to pipette everything in there, or your pipetting error”

Researchers must adapt their position in order to facilitate a particular process, such as having the fingers outstretched next to each well of a 96-well plate to make sure they introduce the solution needed in each well. As such, the ways in which they react to the requirements of the procedures they perform is their choice. The object does not impose a particular reaction.

Scientists’ concerns emphasize one major common aspect, namely, their affirmation of their own agency by emphasizing their duties to follow the protocols, plan ahead, make correct measurements, keep the laboratory clean and position instruments well. In the excerpt below, showing how a technician is instructing a research assistant in molecular work, the

technician is concerned with the consistency of pipetting. Further, for her, the convenience of using particular pipette tips is important. In the case of molecular work, as tools have a fairly narrow possibility of usage, the choice is between different brands of the same tool, as different brands have different properties.

Lili, the technician, takes the rack with the samples, and tells Mike, the research assistant, to arrange the plates on the bench: the master-plate in the middle and three empty plates in a column to the right and three to the left of the master-plate. She tells him to take a multichannel pipette and set it to 2 microliters. She goes to the shelf with pipette tip boxes above the PCR machines and takes a box of tips that looks different than the boxes she uses normally. "When you are making plates, take these boxes, because they are better." She tells Mike. Now she gives him instructions on how to pipette from the master-plate to the other plates with the multichannel: "Make sure that the DNA is in all tips... Don't change the orientation; that will mix the samples." Then she tells him to mix the DNA solution, since it hasn't been mixed. I ask her why the other pipette tips are better than the ones she normally uses. She says that every manufacturer has a different quality. Lili gives more instructions to Mike: "Go all the way to the bottom and then release, so that it's not stuck to the wells." She goes on with the explanation about pipette tips: you can use the special pipette tips multiple times, about 6 times without changing them, whereas you can only use the other tips 2 or 3 times. She says that pipetting error increases the more you pipette. The pipette will take less quantity the next time you pipette. I ask her why that is the case. She speculates that it may be

because of the plastic, but doesn't know for sure. She ends: "I always want the best things, because they make my life easier".

This excerpt illustrates the perception of some objects in the molecular lab, plates, pipettes, pipette tips, as non-agents. The technician frames all the instructions she gives to the apprentice in a way that emphasizes the control the research assistant has over each aspect of the tasks in the protocol. The apprentice is to take the boxes with better pipette tips, position the plates in the correct way to avoid mixing samples, mix the DNA solution and avoid getting DNA stuck to the wells. Any resistance from all these tools, or any action that would prevent these steps from taking place properly, would be the apprentice's fault. The technician's language does not reinforce the agency of fly DNA. DNA gets stuck to the wells simply by its physical properties and small dimensions. Although it is specifically these characteristics that would lead Latour for instance to claim the agency of DNA, they do not seem constitutive of agency for the technician.

Having knowledge of each protocol step is important for performing the work in the molecular lab and illustrates researchers' affirmation of their agency. Scientists can manipulate protocols, if they know the consequences of changing the protocol. Some more experienced and knowledgeable scientists can even contribute creatively to the process. One day I was watching one of the senior researchers extracting fly DNA. She had to separate the DNA from the rest of the cell contents. Her tools, a plastic rack with plastic slim tubes called "columns", were inside the fume hood. The fume hood is a special bench for working with

and discarding dangerous substances. It has metal walls on each side and a pane sliding vertically in the front.

There are 3 columns positioned in a special blue rack in the fume hood. The columns contain silica membranes that need to be moistened with an equilibration buffer to help DNA bind. The contents of three tubes, DNA, enzymes and buffers, must be poured through each of the columns in a specific order. Anne brings three disks of filter paper, which she folds into cones so that they fit in the columns. She says about the filters that they are “a trick I added on my own” in order to trap the proteins so that they don’t clog the columns.

In the context of the quote above, knowledge of the physical and chemical processes that take place between containers and substances guides the scientist’s decisions about the kinds of tools in the lab she can use to manipulate the protocol. By making these decisions she affirms her agency. Furthermore, the researcher seems to treat DNA, as well as all other tools as objects, not agents.

While workers in the molecular lab can have a good understanding of the rationale behind different steps in the protocols they follow, they may at the same time not know all the technicalities behind the way particular components are obtained by the companies that produce various protocols. The companies are protective of their patents, so there is limited information on their products. One day I asked the same senior researcher to explain what barcodes and adapters are and how they attach to the DNA. She explained that they are sequences that the producer of the preparation kit provides, and they are used in sequencing for generating clusters of different DNA samples. When I asked questions requiring very

detailed answers, she admitted: “This is all technology beyond us; I don’t understand it”. This is a dimension of laboratory work that suggests some lab objects are black boxes¹ (Latour 1987) for the laboratory staff². The issue of the behavior of DNA in its interaction with barcodes and adapters only poses problems for the question of agency, however. Put in the right conditions, flies’ DNA combines with these smaller complementary sequences artificially produced. This allows for placing side by side different DNA samples from different flies which, at the time of sequencing can be recognized as distinct. In this whole process, machines are providing the favorable environment for the union to happen, “[b]ut what is measured here? Is it still a biological function or is it just a chemical process?” (Rheinberger 2010, 225). By using the construction “technology beyond us” to refer to this process the researcher appears to subscribe to this uncertainty. Nevertheless, she does not seem to treat DNA as an agent.

Molecular work often involves tool breakdown. The unpredictable behavior of machines brings another important point in the discussion of agency. Researchers depend on the quirks of their machines and need to identify and adapt to them:

Technician: “So *centrifuges are very finicky*. And sometimes during these plate extractions, usually I never had a problem spinning at 6000 G, but nowadays uh, the plates start cracking up, so you lose your samples, and when you call tech support they say you cannot go above a

¹ Latour uses black boxing to refer to products of sciences received as ready-made.

² However, scientists do control the artificial strands. In spite of their limited knowledge about them, they have access to a service that can help them understand how to troubleshoot if they encounter problems.

certain speed. But earlier we didn't have a problem. It was the same manufacturer, the same kind of plate, but they say we had the same recommendations even earlier."

From the way the technician talks about the centrifuge it appears she is endowing it with agency, since it is "finicky", and behaves in ways it did not behave before.

A different way of endowing standard instruments with agency is by naming them. Both molecular rooms of the lab had PCR machines with names. Naming machines is probably what past workers in the molecular lab did for amusement (in one lab the machines were named after the Teenage Mutant Ninja Turtles). However, it suggests the importance these machines have in the lab. Several processes (library preparation, DNA cloning etc.) require the use of PCR machines; as such, these machines are used constantly. By naming them, scientists indirectly confer them a sort of identity, or agency. As such, scientists make machines participate in making the social.

The manner in which scientists formulate their concerns, by positing themselves, or their apprentices as the subjects of their sentences, emphasizes their status as agents in the interaction with their tools. In practice, scientists treat their instruments as non-agents; however, at instances in their speech, for example when they talk about difficulties encountered from machines, or by naming certain machines, they seem to afford instruments agency.

As far as RNA/DNA samples are concerned, the researchers seem to treat them as non-agents, both when talking about them, as well as when handling them. Nevertheless, the question of the agency of DNA/RNA stands out as needing more investigation, given the

uncertainty emphasized by Rheinberger regarding the processes which DNA/RNA undergo in the lab.

CHAPTER 7 AMBIVALENCE AND DATA ANALYSIS

In statistical work, flies are data points, that is, symbols. With statistical work the fly reaches its final destination in its metamorphosis in the lab. As in fly work and molecular work scientists tend to display ambivalence in their speech towards the software used. I did not conduct sufficient participant observation with scientists doing analyses to be able to generalize conclusions to the whole lab. The interviews suggest scientists take responsibility regarding the use of software. As such, they conceptualize it as non-agent. At the same time, practice and talking about practice suggests some researchers interact with the software as if it were an agent.

The researchers introduce the data gathered on sheets or in their lab books into Excel. Two researchers film fly aggression, so they record their data directly into a special computer software, JWatcher, after watching the videos of fighting flies. For some projects, researchers use the *Drosophila* Activity Monitor, located in a special incubator. The software, directly connected to the monitor, records the data. Eventually, researchers transfer the data in Excel.

The scientists then conduct basic descriptive statistics and inferential statistics in SAS, R or JMP. Every researcher who coordinates a project does his/her own statistical analyses, such as the descriptive statistics (mean, standard deviation etc.), and inferential statistics (ANOVAs, *t*-tests etc.). Two postdocs were doing the GWAS – the phenotype-genotype correlations – at the time of my presence in the lab. Some of the other researchers, mainly PhD students and postdocs tried to learn to use the GWASpi.

In speech, the GWAS specialists emphasize the non-agent dimension of the statistical software and the data. In the software, flies are a collection of data. One of them mentioned in the interview: “After doing that analysis, you start to think of things as just observations, rather than actual fruit flies. So you have a big matrix that you're dealing with and that's all that I think about”. Researchers manipulate flies as data with their statistical tools, as the other GWAS specialist explained: “So statistics is just a tool.” He went on to compare it with the other material tools in the lab, such as vials and dissection scopes.

The kinds of concerns that people in the lab have when doing data analysis typically differ among the GWAS specialists and the project coordinators. The project coordinators' concerns typically are doing the right analysis and writing the codes correctly, as they are not experts of the statistical software.

Postdoc: *“One of my biggest concerns, since I am not a statistician: I am doing it correctly? [...] Am I using the right model? Am I using the right test? Am I interpreting it correctly?”*

By asking such questions, researchers stress their agent role in controlling the analyses by making the correct statistical procedures.

The GWAS specialists, as well as other people in the lab who have tried to understand the GWAS Pipeline, are concerned that statistical models produce artificially inflated significance. In this lab, I noticed a tendency to take results with a pinch of salt and think critically about software output:

PhD student: *“The shortcomings of our GWAS, yes of course. I mean there's way too many... There's a plague of multiple testing. We're doing so many little tests [...] So, there's tons of sites in the genome to tests. There's tons of SNPs, but there's a finite number of flies that we can actually measure. And so we're doing all this multiple testing with this relatively small group of phenotype measurements, so then that can artificially inflate [...] the significance of your SNPs; so the things that come up, you can think that they're significant, but they're not actually, it's just an artifact of multiple testing.”*

In her critique of the GWAS conducted in the lab, the PhD student refers to the lack of alignment between several requirements for the procedure using GWASpi. Because of the numerous sites in the genome subjected to comparison, the number or homozygous strains of flies would have to be very high. There are however, only close to 200 DGRP lines that managed to survive the process of inbreeding. This number is however not sufficient. Consequently, repeated testing is required. Researchers must engage in a process of deciding which combination of genes best suits an answer to their questions and constructing the most sensible results. They thus affirm their agency in the interactions with the software.

However, the way that another PhD student talked about her use of SAS software suggests that her practice involves treating the software as agent: “I spend half my time yelling at SAS”. Further, when I was observing one of the senior researchers writing code and doing some simple analyses in SAS, she was constantly murmuring questions and instructions to the software. This was a notable interaction that might be interpreted as the

researcher projecting agency on the software and acknowledging its role in performing the tasks she wants it to perform.

In the data analysis stage, researchers maintain their agent position, while emphasizing in speech the non-agent character of the data and the software used. This attitude seems more prevalent among the lab staff than the contrasting attitude, conferring the software agency in practice, which I only noticed from talking to two researchers. Although the observations referring to agency in the data analysis stage may not be generalizable to the whole staff, it still illustrates a tendency towards ambivalence.

CHAPTER 8 DISCUSSION

To bring some broader insights to the fore, I observe the variations in *Drosophila*'s transformation in the three stages of research considered in this study. Further, I discuss scientists' critical thinking about their work and interactions in the lab, which, I contend, may influence to a high degree their constant affirmation of their own agency. In connection to researchers' ambivalence regarding agency of non-humans in the lab, I highlight the distinct conceptualizations of tool. Overall, these observations about the character of the *Drosophila* lab considered in this study facilitate conclusions relevant to the broader conversation around 'agency'.

Walking through each research stage that involves systematic use of various instrumentations suggests *Drosophila*'s transformation is a non-linear process. This means that within one stage of research, the fruit fly does not have one single form, but two or multiple forms stored in different media. Scientists transform the fruit flies after each behavioral assay from living organisms into scores on a sheet of paper or in computer software. In the molecular stage, the flies transform from lifeless, frozen organisms into DNA or RNA samples. The scientists use various substances and machines to process the DNA/RNA samples into inscriptions of nucleotide sequences. Finally, in the data analysis stage, fruit flies are data points that the researchers convert using software into comprehensive analyses of all the flies studied. The complexity of shapes *Drosophila* takes is relevant for critiquing the use of 'agency' in the current theory to understand lab interactions. That is because humans relate differently to each form flies take.

Given the main conceptualizations of agency in the theory on laboratory interactions (agency as intrinsic to all machines and organisms; agency as a property of a component of a network; agency as conforming to a typology), the ethnographic data from the *Drosophila* lab indicates these theories limit the understanding of the interactions between the researchers, instruments and model organisms in this lab. Robert Hooke's view of agency endowed objects with intrinsic powers that dictated the effect of an object on another object or on an organism. In Actor Network Theory, agency had mainly a methodological function; the social scientist studying a laboratory should take into consideration all humans and non-humans in the network of the lab equally (symmetrically), as all of them are agents of change on one another. Kaptelinin and Nardi took Latour's ideas of symmetry literally and created a typology of conditional, need-based and delegated agency to demonstrate that agents in a network do not have symmetrical positions. I do not engage here with interpreting ANT's claims or the arguments of its critics. Instead I suggest that the first two theories have something in common with respect to the idea of agency: they discount the perspective of the humans engaging with objects and organisms in particular lab settings. The third framework partly considers humans' primacy in that they get to delegate agency to other entities. Nevertheless, it is a complicated framework to apply. Even Knappett and Malafouris' argument for a situated agency does not provide a convincing framework for discussing human-non-human interactions in the lab as it brings forth a standard for defining agency. The humans' perspective is essential, because it shapes in large measure the interaction between these entities.

Observations in the *Drosophila* lab illustrate the hierarchy between scientists, fruit flies and objects. In this hierarchy humans have the most powerful role in making decisions about the objects and organisms with which they interact. The researchers do not see themselves merely as spokespersons of the organisms they study and objects they use, as Latour would have it. Instead, they are responsible for the changes in the lab. Perhaps most important in suggesting humans' control over the equipment and processes in the lab is their critical attitude towards their scientific work. Sitting with one of the technicians (and key informant) in her office I remarked at one point in our discussion that it was very insightful for me to be in a laboratory and learn how science is done. My interlocutor aptly remarked: "not how science is done, but how we do science". For my technician science is something a person does, so it is in interplay with human idiosyncrasies, and more specifically, the idiosyncrasies of the humans in the lab.

Further, the researchers in the lab are aware of the human bias inherent to conducting experiments. One day, as I was conducting my experiments, I asked a senior researcher who had experience with my assay to double check my observations. She leaned in close looking at my assay and told me she cannot help me much because she may have a different judgment than I regarding the behavior of the flies. Moreover, as I illustrated earlier, scientists are aware that GWAS results are not completely reliable. As such, in general, the scientists in this lab have a keen awareness of their own limitations, as well as the limitations of their tools and experimental setups.

In their interactions with and speech about their laboratory environment scientists are ambivalent about the agency of the fruit flies (and the various forms they take) and of the instruments they use. Scientists undermine *Drosophila*'s agency conceptually by perceiving it as a tool. Furthermore, researchers treat material tools (be they sorting tools, PCR machines, or statistical software) mainly as non-agents. However, sometimes they anthropomorphize them, as such conferring agency to them through particular uses of language (referring to them as “this guy”, or giving them names). They do not confer agency to instruments in practice as they do in the case of the flies. It is important to observe the distinct perspectives in relation to “tool” when the term designates objects as opposed to fruit flies. When fruit flies are tools, they are only non-agents.

Importantly, scientists' ambivalence regarding agency in the lab, as well as the different perspectives towards tool are manifested in speech and practice. A pertinent question is therefore: what do we premise, scientists' speech or their practice, since each tells a different story? Indeed, scientists themselves suggest they are not always the primary agents in the lab. Nevertheless, they are the only ones who create meaning in the environment in which they work. The way they do so is by recognizing their fluid negotiations with the other entities in the lab, as well as by imposing their symbols on the inanimate things around them. These dynamics may be different in other laboratories. As such, a story that contains these different perspectives (no matter how contradictory) enables the illustration of the complexity of the lab environment.

CHAPTER 9 CONCLUSIONS

This study of the *Drosophila* laboratory opens a conversation about ethnographic material from the contemporary genetics laboratory by highlighting the issue of non-human agency. Different writings about the laboratory indicate the significance of this concept for understanding lab interactions or for approaching the study of lab interactions. I gathered evidence from scientists' speech and practice to answer questions related to agency as such, without assuming a priori the agency of all the things, organisms and humans in the laboratory. This method proved useful in illustrating that agency is situated in the context of the laboratory. To analyze laboratory interactions it is instrumental to give primacy to scientists' perspectives of their relationship with their laboratory environment.

Theoretical frameworks in the social study and the philosophy of science acknowledge the importance of scientists in defining their relationship with the objects and organisms surrounding them. The notion of indeterminacy emphasizes the importance of human decision making. In all three stages of *Drosophila* transformation scientists stressed the significance of decision making with respect to choosing instruments, designing experiments and interpreting data analysis results. Furthermore, the intersection between instruments and the model organism is important for the researcher to understand more about the instrument he/she uses as well as the model organism. In Chapter 5, which discussed fly work, the researchers gave special attention to the significance of flies' reactions to their instruments (CO₂, the "flydiator arena"). The concept of agency does not pertain to indeterminacy and instrument-experimental object interface as they emerge from theory.

Using the notion of agency with respect to these theories in my study was a strategy to situate scientists' conceptualization of their work within the laboratory.

It is important to mention that the strategy presented here is only useful if the focus is on the interactions within the laboratory. Kleinman (2003) suggests that agency, as well as indeterminacy, are sometimes limiting concepts. He argues that for a deeper understanding of the practice of university science, social scientists should look at structures, rather than at technical matters (Kleinman 2003, 5). Labs operate in a wider system of interconnected public and commercial institutions that provide resources to or take resources from the lab. Nothing in a lab is confined solely to the lab. I find this to be a strong claim that relates to my own observations in the *Drosophila* lab. Acknowledging that agency is defined in the lab through scientists' sociality with the non-humans there brings into focus the importance of community and social structure. This bridges the discussion about the interactions unique to the *Drosophila* laboratory with a potential broader conversation about the extended structure in which this lab is embedded.

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APPENDICES

APPENDIX A: GLOSSARY

Assay: a series of steps in using laboratory instruments for evaluating qualitatively or measuring quantitatively the characteristics of an experimental object.

Buffer solution: a solution used to stabilize the pH of a liquid after dilution.

Cloning: molecular techniques that assemble strands of DNA from different sources which will further be transferred to a host cell.

DNA (deoxyribonucleic acid): a macromolecule in the shape of a double stranded spiral, which encodes genetic information used in the development and functioning of organisms. The double stranded spiral is packed in chromosomes and is housed in the nucleus of the cell. The following alternating nucleotides A, T, G, C form DNA.

Experimental design: refers to the way in which scientists organize their study: sample populations and establish the treatment to which to subject them.

Experimental system: according to Hans-Jörg Rheinberger (1997, 238), is "A basic unit of experimental activity combining local, technical, instrumental, institutional, social, and epistemic aspects."

GAL4/UAS driver insertion: a biochemical method used to study gene expression and function in organisms such as the fruit fly.

Genotyping: the process of determining differences in the genetic make-up (genotype) of an individual by examining the individual's DNA sequence and comparing it to another individual's sequence. Genotyping reveals the alleles an individual has inherited from its parents.

GWAS (genome-wide association study): is an examination of many common genetic variants in different individuals to see if any variant is associated with a specific trait. GWAS typically focus on associations between single-nucleotide polymorphisms (SNPs) and physical or behavioral traits.

GWASpi (GWAS Pipeline): a "user-friendly, multiplatform, desktop-able application for the management and analysis of GWAS data" (Muñiz-Fernandez, Carreño–Torres, Morcillo-Suarez and Navarro, 2011, 1871)

Library: a collection of DNA fragments that is stored and propagated in a population of micro-organisms through the process of molecular cloning.

Metabolic pathway: a series of chemical reactions within a cell.

Microarray: a collection of microscopic DNA spots attached to a solid surface. It is used for measuring the expression levels of a large number of genes or to genotype different parts of the genome.

Minos insertion: another type of insertion with transposable element (a DNA sequence that can change its position in the genome of an organism and disrupt other genes)

PCR (polymerase chain reaction): a biochemical technology used in molecular biology to amplify a single or several copies of a piece of DNA and produce thousands to millions of copies of a particular DNA sequence.

P-element insertion: the introduction of a genetic sequence, called transposon, which can copy itself at a particular site in the fly genome and disrupt the expression of a gene.

Phenotyping: phrase used by people in the lab to describe the process of observing physical or behavioral traits in fruit flies.

Protocol: the set of procedures that the scientist must follow in the implementation of an experiment. There are protocols both for behavioral assays and for molecular work.

RNA (ribonucleic acid): the complementary replica of one strand of DNA that is transported outside the nucleus and is translated into amino acids which form proteins.

RNAi insertion: a technique that introduces in the fly a gene that expresses a type of RNA that disrupts expression of another gene. It forms a hair-pin structure around the messenger RNA of a gene and degrades it.

RT-PCR (reverse transcription polymerase chain reaction): a variant of polymerase chain reaction (PCR). This technique is used in molecular biology to detect RNA expression levels. RT-PCR is often confused with real-time polymerase chain reaction (qPCR) by students and scientists alike.

SNP (single nucleotide polymorphism): a DNA sequence variation that occurs when a Single Nucleotide — A, T, C or G — in the genome (or other shared sequence) differs between members of a biological species or paired chromosomes.

Transgenic (or genetically modified) organisms: contain in their genome (in body cells, as well as sex cells) a foreign DNA fragment (from a different organism) and are able to pass it on to their offspring (Houdebine 2005)

APPENDIX B: INTERVIEW GUIDE

- 1) How long have you been working in this laboratory?
- 2) What kinds of experiments do you work on mostly?
- 3) What kinds of equipment do you use most often for those experiments?
- 4) What are three key concerns that you have when you phenotype fruit flies?
- 5) What are three critical concerns when you do molecular experiments?
- 6) What kinds of difficulties caused by the equipment do you encounter during experiments?
- 7) Do you have an instrument or piece of equipment you prefer? Why?
- 8) Why is it useful to do research on transgenic *Drosophila*?
- 9) What is the importance of modeling of *Drosophila* traits and behavior? What are three essential concerns in relation to modeling?
- 10) What are the shortcomings of statistical modeling?