Drawing and Visualisation Research

Published in *TRACEY* | journal
Thinking
December 2014

www.lboro.ac.uk/departments/sota/tracey/
tracey@lboro.ac.uk

Special Edition:
Drawing in STEAM

A CYCLE OF DRAWING IN RESEARCH INTO LIFE AT THE MOLECULAR LEVEL

Kathryn Garner [a]

[a] School of Clinical Sciences, University of Bristol. info@kathryngarner.co.uk.

At the nano-scale, our body cells are packed with millions of tiny protein machines, each with a key part to play in human physiology. The understanding of what each protein does and how it interacts with its neighbours is fundamental in keeping us well, fighting disease and designing new, more effective drugs. The more questions we ask, the more complex life at this molecular level appears to be. Despite improving technologies, the practice of drawing remains an integral part of the discovery process, allowing the maker to think through and bring together distinct ideas, analyse protein-protein interactions and plan ways to test hypotheses. In this paper, I discuss how a cycle of drawing is used, with each drawing type feeding forward into the next, and each iteration improving clarity and the collective knowledge. Sketches and drawings made to understand the wider scientific literature inform the planning of experiments and weekly timetables, the results of which feed back to embellish what was initially understood. I identify a trend at all stages of the cycle, where drawings begin life as transient objects, becoming more permanent as the thought contained within them persists. Permanence frequently involves the digitalisation of the drawing, allowing increased viewing through dissemination to the wider community in journal articles and discussion at scientific conferences.

INTRODUCTION

There are an estimated ten trillion cells in the human body, organised into tissues specialised for particular functions. Each tissue or population of cells must carry out its job, such as absorption of nutrients into the body in the case of the small intestine, together with its house-keeping functions of energy production and cell renewal. Just as one member of an orchestra must not play their part without regard for the tune played by their colleagues, a single cell must communicate with neighbouring cells and with cells in other tissues to ensure that they are carrying out their specialised role exactly in line with the needs of the body as a whole. Communication is achieved by the production of small molecule messengers, such as hormones like insulin, adrenaline and cortisol. These are synthesised and secreted on demand, and travel around in the blood, searching for the appropriate target tissue to pass their message on to. A key will only fit into a certain shape of lock; the hormone will only be recognised by a particular receptor found on the surface of its target cell. Binding of the messenger molecule to its cognate receptor sets in motion a series of inter-molecular interactions, which communicate the message into appropriate compartments in the interior of the cell. Any misinterpretation can have disastrous consequences, and it is therefore essential that we understand how these communication pathways operate so that we can find ways to fix them in disease states, including cancer.

Understanding how the protein machines work and interact with one another would be impossible without the use of drawing to aid in the thought process. Drawing enables the thinker to bring together consideration of different scales, from the atomic to the whole tissue and organ level, and to imagine complex processes that are not able to be visualised using microscopy or other imaging methods. The use of drawing as a teaching tool in the classroom is widely acknowledged (Dempsey and Betz, 2001; Driver et al. 1994; Lowe, 1987; Nyachwaya et al. 2011), yet its use as a thinking and communication tool by the researchers themselves has not been reported. In this paper, I discuss the concept of a drawing cycle underpinning all research methods in molecular biology. The first part of the cycle involves placing the research question into context through regard for the wider scientific literature. These drawings inform the next part of the cycle: planning experiments to test hypotheses in light of what is already known. The results of new experiments supplement the existing knowledge, bringing new questions to the forefront, and so too the planning of new experiments. Each stage in the cycle employs the tool of drawing.

THE DRAWING CYCLE IN BIOMOLECULAR RESEARCH

The role of drawing in understanding the wider context

In order to begin to find ways to answer a particular research question, it is imperative that one first has a good understanding of what came before. Put simply, there is no point in repeating work that has already been done. In reading about the research that has been attempted, the scientist might learn methods or important points that they had not thought of, including experiments to be avoided for one reason or another. Scientists report their findings through publication in peer-reviewed journals, through talks and poster presentations at conferences, and discuss methods in online forums. The use of images and diagrams is key in this communication of information. Drawings allow the rapid integration of findings from different sources, and through the use of symbols and accepted jargon can be easily copied and modified as they are passed from one researcher to another. An example drawing is shown in Figure 1A, which is a sketch of a diagram in a review article about signalling pathways in blood platelets. The top part of the drawing describes several pathways by which a signal is transmitted from outside the platelet to the inside though physical interactions, conformational changes and enzymatic reactions. Towards the lower half of the drawing, the portrayal of the signal becomes less detailed, stating instead the wider phenotypic consequences of the interactions.

In all parts of the cycle, drawings begin life with a sense of immediacy, made using whatever tools and surfaces are available to hand. This may be pencil or pen on paper, but may extend to the back of a catalogue, the corner of a newspaper or a napkin. The importance is in getting the idea down quickly before it is forgotten. Often, a quick sketch made during a conversation might aim to establish that two researchers are thinking along the same lines. Dry-wipe markers on a whiteboard might be employed for drawings made in a communal setting, such as during a brain-storming session in a lab meeting (Figure 1B). Here, the scientists have discussed the inter-conversion of members of the phospholipid class of lipid from one form to another, with respect to the location of enzymes required within the cell. The drawing includes hypotheses about unknown connections and how one might target each part by experiment. In general, little care is given to the colours or scale used, and much is gained through the experience of drawing and discussion. Often a change in colour marks a pause in making the drawing (perhaps minutes, hours or more). A new colour might have been picked to introduce new ideas to an existing drawing, or because the original colour has disappeared or expired. In this way, the use of multiple colours might reflect the time taken in the communal thinking process for the evolution of ideas.

These initial drawings have a tendency to be transient in nature: markings on a white board are easily wiped away; a drawing on a scrap piece of paper is easily lost or discarded. A drawing in a notebook or lab book may also be considered transient as it might only be happened upon as long as that book is in use. When that notebook is finished and a new

one started, the drawing might only persist if it is re-drawn. A drawing on a whiteboard is less transient than a drawing in a notebook in some ways, since it has the opportunity to be seen constantly by all that pass it. In the same way, a drawing on a piece of scrap paper may be pinned on a wall and remain in the researchers' consciousness until it is covered up or removed. All of these types of drawing may be erased and surpassed by a new drawing or theory. Further modification and refinement is required to prolong its life and ensure it remains in the researchers' thinking. This is frequently achieved using digital methods, which also enable the communication of the thought to the wider scientific community. This dissemination might occur through publication in a journal or grant application for research funds, reproduction on a poster, or in a slide in a talk at a scientific conference.

The use of computer software not only makes a drawing more permanent, but also allows the processing of a large quantity of complex data into dynamic drawings that can be more easily understood and manipulated. This includes complex interaction networks, such as those generated using GeneMANIA interactive software (Mostafavi et al. 2008) (Figure 1C), and the structure of the individual proteins themselves (Figure 2). The protein shown in this latter figure is RdgBβ, which transfers phospholipids between different intracellular membrane compartments. The computation of the positions of individual atoms making up a protein in three-dimensional space is not possible using the human mind alone, and so computers aid us in this task. Software such as the PyMOL Molecular Graphics System allows the rapid rendering of a molecule, permitting the researcher to try different ways of seeing the structure in order to determine the best method for visualisation. The structure can be manipulated in three-dimensional space, which is an essential requirement for understanding and thinking about how these tiny protein machines work. Theories about the importance of particular amino acids in protein-protein interactions or enzyme functions can be formed and later tested by experiment.

The visualisation of protein structure using computer software can feed back into quick, hand-drawn sketches. The X-ray crystal structure of RdgBβ has not been reported to date, and so the protein's amino acid sequence has been modelled on a reported structure of another member of the transfer protein family (Figure 2). RdgBβ is distinct among this family as it carries a long amino acid extension or 'tail'. Bioinformatics programs indicate that this tail lacks a formal structure, and therefore in Figure 2D this part of the protein has been added in red by hand using Adobe Illustrator, below the computer-generated core structure. Often, the researcher uses the computer-generated form as a starting point to simplify the structure into an easily reproducible, symbol-like form.

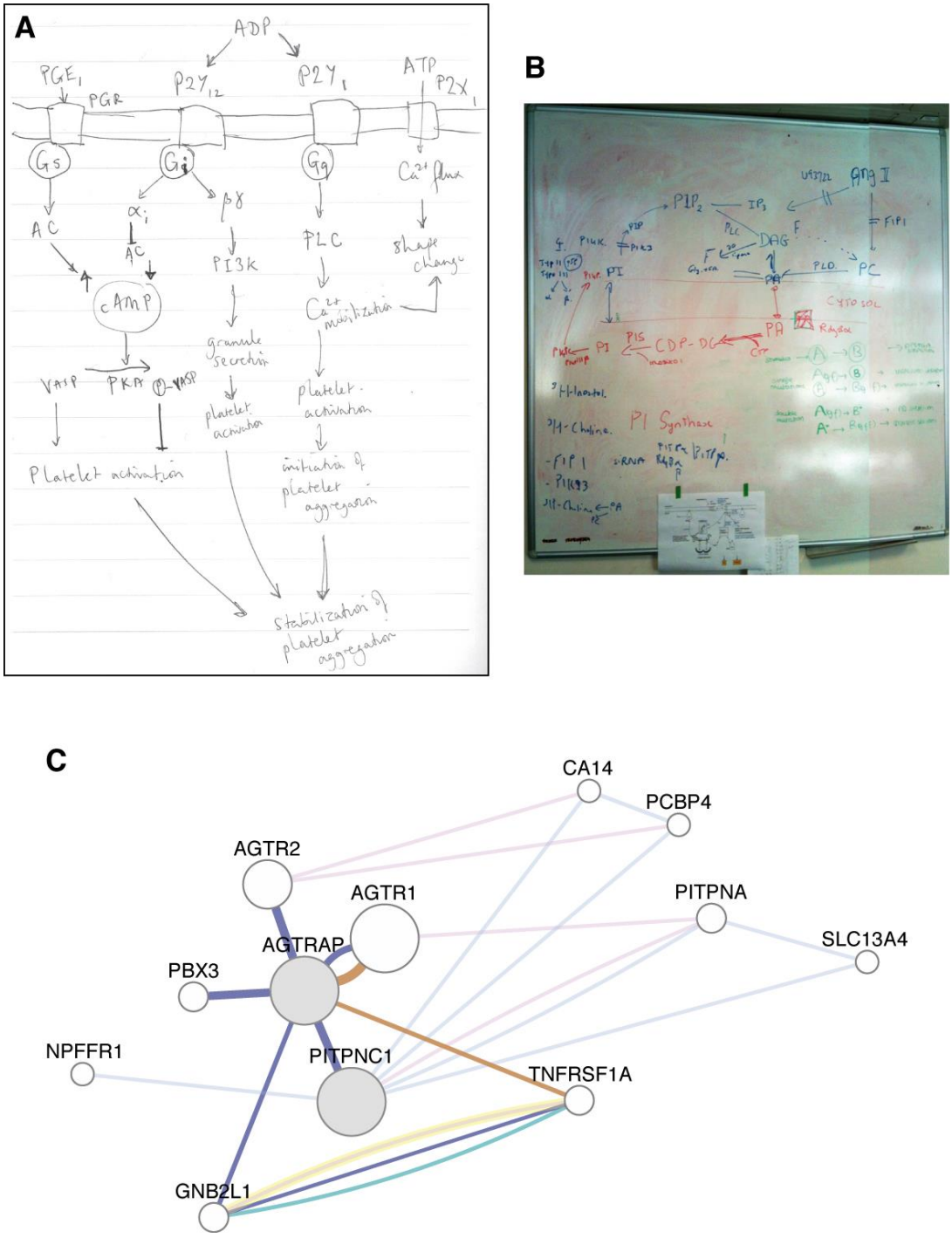


FIGURE 1: PATHWAY DRAWINGS. **(A)** SKETCH MADE FROM THE SCIENTIFIC LITERATURE OF SIGNALLING PATHWAYS DOWNSTREAM OF BLOOD PLATELET CELL SURFACE RECEPTORS INVOLVED IN PLATELET ACTIVATION AND AGGREGATION FOR BLOOD CLOT FORMATION. BLACK BIRO ON NOTEBOOK PAPER. **(B)** GENERALISED LIPID SIGNALLING NETWORK WITH COMPOUNDS THAT CAN BE USED TO TARGET PARTICULAR PARTS OF THE PATHWAY. COLOURS USED NOT SIGNIFICANT AND MAY HAVE BEEN CHOSEN MERELY FOR VARIATION. DRY-WIPE MARKER ON WHITEBOARD. NOTE ALSO DIGITAL DRAWING OF A DIFFERENT PART OF THE PATHWAY FIXED TO THE BOTTOM OF THE BOARD. **(C)** INTERACTIONS INVOLVING AGTRAP (ATRAP) AND PITPNC1 (RDGBB) PROTEINS, GENERATED USING THE GENEMANIA INTERACTIVE SOFTWARE (MOSTAFAVI ET AL. 2008). WEB NODES CAN BE DRAGGED BY THE USER TO CHANGE THE NETWORK SHAPE OR EMPHASIS, AS REQUIRED. PURPLE-BLUE, PHYSICAL INTERACTIONS REPORTED; PINK, PROTEINS HAVE A COMMON PATTERN OF EXPRESSION; ORANGE, INTERACTION PREDICTED BY COMPUTATIONAL METHODS; TURQUOISE, PROTEINS HAVE BEEN REPORTED TO LIE IN THE SAME PATHWAY; PALE BLUE, PROTEINS CO-LOCALISE.

In the next figure, the RdgBβ structure has been simplified so that the core domain is represented by a rough lozenge-shape (Figure 3A), or hexagon containing a phospholipid (Figure 3B). This simplified structure retains its long tail, characteristic of this protein. Using this process, a large dataset (the co-ordinates of atoms in space) is made into a drawing by computer software that produces images in response to a defined set of rules. This has the potential to uncover information about individual proteins, perhaps providing clues about the location and characteristics of potential interaction surfaces. This may then feed forward to imaginative drawings and the design of new experiments, which have the potential to produce further large, unwieldy datasets.

The design of new experiments

Armed with a broad understanding of the scientific literature and bursting with new theories to test, the next stage in the research process is the experimentation itself. Drawing is key here too, although those made are more likely to remain on scraps of paper or hidden in lab books to be viewed only by their maker.

Whereas drawings made whilst understanding what has come before could also be referred to as diagrams, those made whilst thinking about and planning experiments may also be thought of as maps. These may be maps of activity or time over a day, week or month, detailing several experiments at different stages, including preparation and data analysis. They may also be the maps of the experiments themselves: the layout of test-tubes in a rack, or conditions on a cell culture plate (an example of the latter is shown in Figure 4). Drawings of this type ensure the experiment is properly planned with the correct controls before time and resources are spent. They also allow the experiment to be set up in such a way that will minimise errors, reagents and laboratory ware. Lastly, when many experiments have been carried out and the outcome known, maps of figures to be made for publication might be produced, arranged in such a way so as to convey one's findings as clearly as possible.

The results of the new experiments may next be used to modify or confirm predictions made in the early part of the drawing cycle. They may cause rough, transient drawings to be discarded or forgotten. Alternatively, new results might lead to particular drawings and ideas increasing in permanence by digitalization methods or by fixing to a wall. The scientific literature might again be consulted, in light of new results. These new results might themselves be published, further contributing to the greater knowledge. From here, new theories can be conceived, new experiments planned, and so the cycle of drawing in biomolecular research continues.

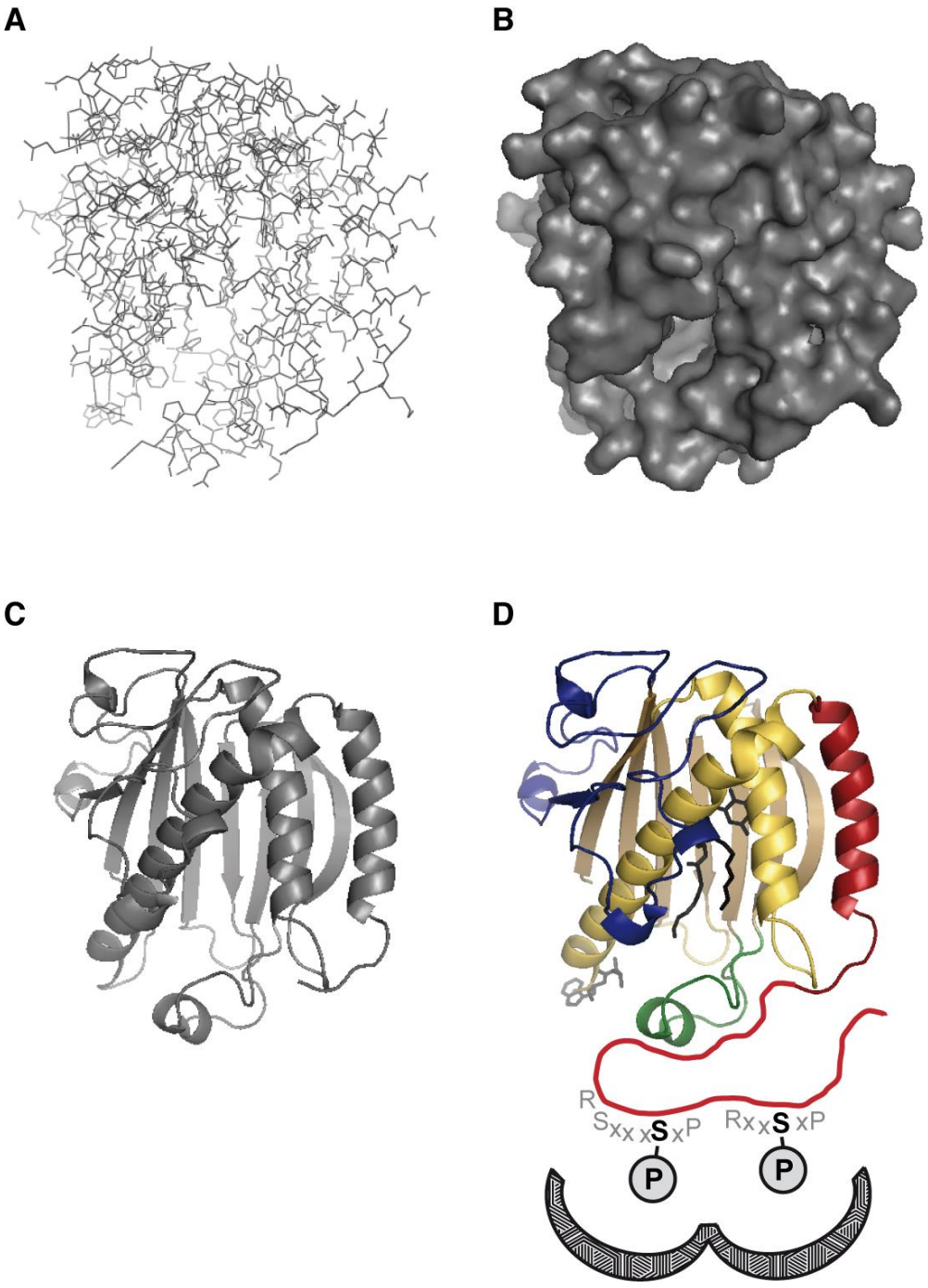
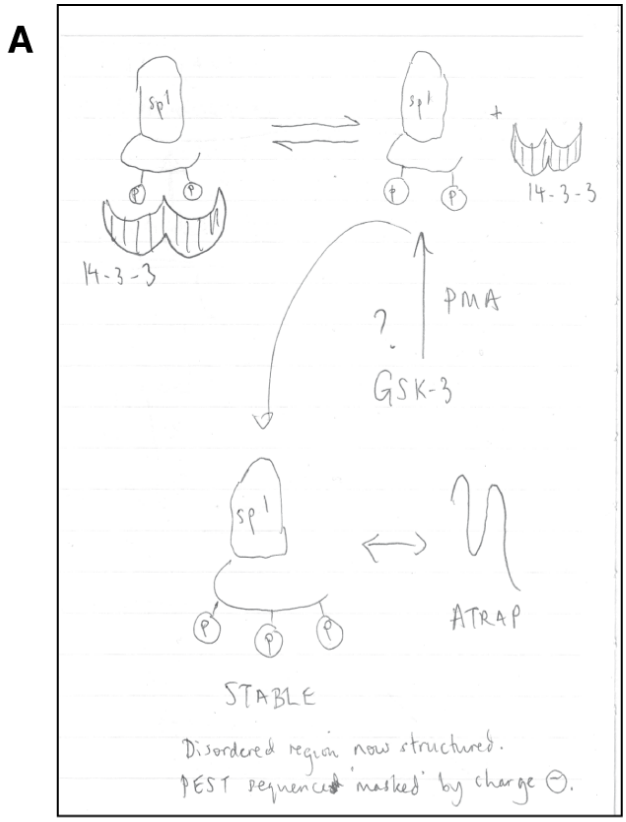


FIGURE 2: DRAWINGS DESCRIBING PROTEIN STRUCTURE. RDGBB STRUCTURE MODELLED ON THE X-RAY CRYSTAL STRUCTURE OF PITPA BINDING PHOSPHATIDYLCHOLINE (PDB: 1T27; (YODER ET AL. 2001)) USING MODELLER SOFTWARE (ESWAR ET AL. 2006). STRUCTURE VISUALISED USING PYMOL MOLECULAR GRAPHICS SYSTEM, VERSION 0.99, SCHRÖDINGER, LLC. (A) BONDS BETWEEN ATOMS IN THE PROTEIN STRUCTURE SHOWN AS LINES. (B) MOLECULE RENDERED TO SHOW WHAT THE SURFACE OF THE PROTEIN LOOKS LIKE. (C) PROTEIN STRUCTURE SHOWN AS A CARTOON WITH A-HELICES AND B-SHEET USED TO SHOW THE DIFFERENT CONNECTIVITY BETWEEN THE PROTEIN RESIDUES. (D) CARTOON STRUCTURE COLOURED TO HIGHLIGHT DIFFERENT PARTS OF THE STRUCTURE: RED, AG-HELIX; BLUE, REGULATORY LOOP; GREEN, LIPID EXCHANGE LOOP. PHOSPHATIDYLINOSITOL LIGAND HAS BEEN SUPERIMPOSED INTO THE BINDING POCKET AND IS COLOURED BLACK. FURTHER ELEMENTS HAVE BEEN ADDED IN ADOBE ILLUSTRATOR: LONG, DISORDERED C-TERMINAL REGION (RED); R, S, P DENOTE AMINO ACIDS FORMING A CONSENSUS SITE FOR 14-3-3 BINDING. LETTER P IN A CIRCLE STANDS FOR A PHOSPHATE MOIETY, WHICH MODIFIES EACH OF THE SERINE (S) RESIDUES IN THE PROTEIN TAIL AND IS REQUIRED FOR 14-3-3 BINDING (GARNER ET AL. 2011). DIMERIC CUP-SHAPED STRUCTURE AT THE BOTTOM OF THE DRAWING SYMBOLISES THE 14-3-3 PROTEIN.



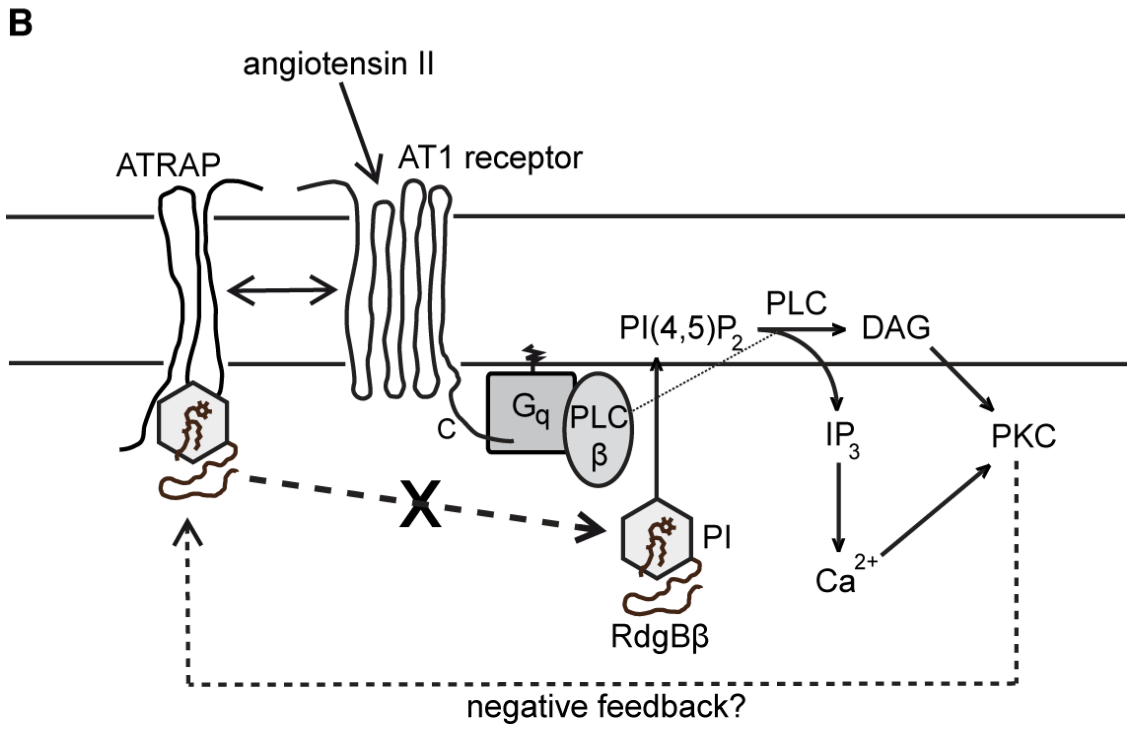


FIGURE 3: SCHEMATIC DRAWINGS MADE IN AN EFFORT TO UNDERSTAND PROTEIN FUNCTION. (A) BLACK BIRO ON NOTEBOOK PAGE. (B) DIGITAL DRAWING MADE USING ADOBE ILLUSTRATOR SOFTWARE. ILLUSTRATION FROM POSTER PRESENTED AT *SIGNALLING 2011: A BIOCHEMICAL SOCIETY CENTENARY CELEBRATION*, UNIVERSITY OF EDINBURGH, UK, 8-10 JUNE 2011, AND *2ND BHF FELLOWS DAY*, QUEENS' COLLEGE, CAMBRIDGE, UK, 4-5 APRIL 2011.

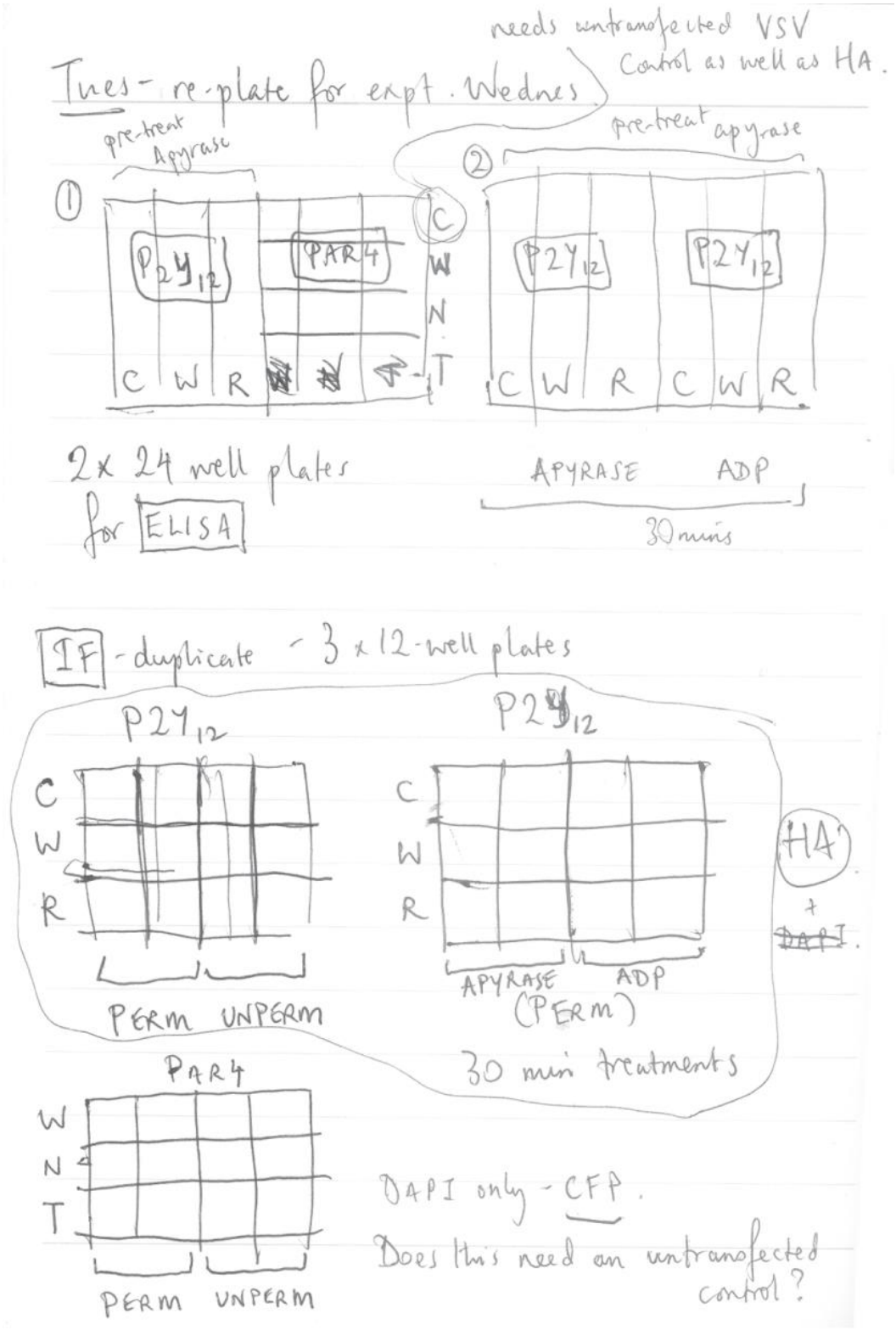


FIGURE 4: DRAWING IN EXPERIMENT PLANNING. RECTANGULAR GRIDS DENOTE 12- OR 24-WELL CELL CULTURE DISHES, AND ASSOCIATED ANNOTATIONS INDICATE POSITION OF PARTICULAR REACTION CONDITIONS IN ENZYME-LINKED IMMUNOSORBENT ASSAY (ELISA) AND IMMUNOFLUORESCENT (IF) MICROSCOPY EXPERIMENTS. BLACK BIRO ON NOTEBOOK PAGE.

CONCLUSION

It is clear that the activity of drawing plays a key role in every stage of the research process, from digesting the wider scientific literature and conceiving new hypotheses, planning time and experiments, to the integration of experimental results within the existing framework of understanding. In this way, a cycle of drawing is undertaken, with the results of new experiments informing the next drawing cycle. Drawings begin as transient objects, being made more permanent should the theories and processes they propose persist.

REFERENCES

Dempsey, B.C. and Betz, B.J. (2001). 'Biological Drawing A Scientific Tool for Learning'. *The American Biology Teacher*, 63, pp.271–281.

Driver, R. et al. (1994). 'Constructing scientific knowledge in the classroom'. *Educational Researcher*, 23, pp.5–12.

Eswar, N. et al. (2006). 'Comparative protein structure modeling using Modeller'. *Current protocols in bioinformatics / editoral board, Andreas D Baxevanis [et al]*, Chapter 5, p.Unit 5.6.

Garner, K. et al. (2011). 'The phosphatidylinositol transfer protein RdgBβ binds 14-3-3 via its unstructured C-terminus, whereas its lipid-binding domain interacts with the integral membrane protein ATRAP (angiotensin II type I receptor-associated protein)'. *The Biochemical journal*, 439(1), pp.97–111.

Lowe, R.K. (1987). 'Drawing out ideas: A neglected role for scientific diagrams'. *Research in Science Education*, 17, pp.56–66.

Mostafavi, S. et al. (2008). 'GeneMANIA: a real-time multiple association network integration algorithm for predicting gene function'. *Genome biology*, 9 Suppl 1, p.S4.

Nyachwaya, J.M. et al. (2011). 'The development of an open-ended drawing tool: an alternative diagnostic tool for assessing students' understanding of the particulate nature of matter'. *Chemistry Education Research and Practice*, 12, p.121.

Yoder, M.D. et al. (2001). 'Structure of a multifunctional protein. Mammalian phosphatidylinositol transfer protein complexed with phosphatidylcholine'. *The Journal of Biological Chemistry*, 276(12), pp.9246–9252.