2025/2026 Science Fair Logbook

Student Names: Katie W & Kiana W

Project Title: Reducing Allergenic Pet Dander Proteins with Eco-Friendly Household Methods: Heat, Vinegar, and Detergent

School: Independent Entry

Purpose & Background: This logbook was primarily written and maintained by Katie W., with both of us (Katie and Kiana) contributing to experimental work, observations, and reflections. First-person singular ("I") represents personal reflections or planning by Katie W., while first-person plural ("we") reflects shared experimental work and decision-making. Entries are chronological and organized into:

- Activities: Tasks completed
- Observation: Notable findings
- Reflection: Why it matters
- Learning: Skills and knowledge gained
- Next Steps: Plans for the next stage

Note - Updates were recorded digitally in my iPhone notes app for convenience and chronological tracking and then pasted into word doc at end for printing & uploading.

<u>November 2025 – Idea Generation and Planning</u>

Activities:

- Reviewed CYSF rules and the judging criteria. Created timeline of events and deadlines. Created a draft schedule for my project.
- Brainstormed project ideas, focused on animal science and allergies
- Emailed school to ask about participation and support
- Began initial research on animal dander and immunotherapy

Observation / Reflection:

- I unfortunately have bad pet allergies. This project combines my personal experience with scientific curiosity. Managing my allergies (through medications, inhalers, and limiting pet access) gives me unique motivation and insight into why this topic is important.
- My motivation: This project has the potential to make pet ownership safer and more enjoyable for people with allergies, which is personally motivating and socially impactful.
- Recognized that the project must be achievable at home in case my school does not approve my request to participate.

Learning:

- Early planning helps identify realistic, impactful projects
- Personal experiences often inspire the most meaningful scientific questions

Next Steps:

- Narrow down project ideas

- Begin background research
- Prepare for potential independent CYSF registration

December 4, 2025 – Independent Registration Preparation
Activities:
- School confirmed no participation
- Began independent registration with my parent's support
- Continued research on allergenic proteins, immunotherapy, and protein denaturation

Observation / Reflection:
- Taking responsibility for independent registration is a lot of work but also feels empowering and highlighted the importance of planning
- My personal allergy experiences motivated me to find practical solutions for people with pets or that would like to have pets

Learning:
- Real-world challenges help refine project range and approach

Next Steps:
- Finalize project idea
- Plan experimental logistics

December 12, 2025 – Team Formation
Activities:
- Convinced my younger sister (Grade 8) to join my project
- Completed initial registration steps with CYSF

Observation / Reflection:
- Working with my sister will make the project collaborative and more enjoyable
- Ethical approval for animal hair collection was emphasized, even though our samples will be collected during routine grooming

Learning:
- Collaboration strengthens planning and efficiency

Next Steps:
- Finalize project title and experimental plan
- Collect baseline fur samples

December 20–24, 2025 – Background Research & Hypothesis Development
Activities:
- Defined the primary research goal: to investigate whether eco-friendly household treatments can reduce allergen-related proteins on animal fur using methods that are inexpensive, accessible, and safe for both humans and animals
- We hypothesized that eco-friendly household treatments that disrupt protein structure would reduce extractable protein levels on animal fur more effectively than distilled water alone, with vinegar producing a moderate reduction due to pH-based denaturation.

- Reviewed online scientific and educational sources, including articles, instructional videos, and laboratory demonstrations related to protein structure, protein denaturation, and the Biuret protein assay
- Explored potential treatment methods, including UV light, household cleaning products, and commonly used eco-friendly practices
- Evaluated each potential treatment for scientific plausibility, safety, accessibility, and real-world household application

Observation / Reflection:

- During our research, we found demonstrations of protein denaturation, primarily using egg proteins, but we could not find studies or experiments that measured protein levels directly on animal fur. This suggested that our project addresses a mostly unexplored question especially at the high school level.
- Initially, we hypothesized that UV light would be the most effective treatment because we use a UV retainer cleaner at home that claims to kill microorganisms. I imagined that a similar device could potentially be adapted to a vacuum or pet grooming tool.
- After contacting the product manufacturer and searching for technical specifications online, we determined that consumer-grade UV devices lack clear intensity data and are likely insufficient for causing meaningful protein denaturation.
- Based on this evidence, we decided to shift our focus toward heat, detergent, and vinegar, which have potentially stronger mechanisms for disrupting protein structure and are safer and more practical for household use.
- We replaced UV treatment option with a low heat method

Learning:

- Protein structure can be altered through multiple mechanisms, including thermal denaturation, disruption of hydrophobic interactions by detergents, and pH changes caused by acids
- A strong experimental design requires evaluating not only theoretical effectiveness, but also safety, feasibility, and reproducibility

Next Steps:

- Collect animal fur samples using ethical and non-invasive methods
- Design controlled treatment procedures for each household method
- Identify safe, accessible tools for quantifying protein concentration

<u>December 21–23, 2025 – Fur Collection</u>

Activities:

- Collected fur from:
    - Bella (short-haired tortie): affectionate, loves being brushed, sheds surprisingly large amounts
    - Duke (long-haired ragdoll): enjoys brushing but only for short periods, sheds moderate amount
- Received small bags of dog undercoat and horse tail hair from family friends
- Browsed online to determine what tools and materials we can easily purchase through amazon or other local stores

Observation / Reflection:

- Bella's comfort made brushing and sample collection easy
- The contrast with Duke showed how behavior and fur type influence shedding
- Realized we likely don't have enough dog / horse hair for many samples, limited dog/horse hair received so far will impact the number of replicates for these sample types and could impact experimental consistency

Learning:

- Fur characteristics affect protein content and treatment outcomes
- Observing the animals' comfort ensures ethical collection

Next Steps:

- Label and weigh fur for baseline data
- collect each animal's photo and details for sample documentation

<u>December 26, 2025 – Experimental Design, Safety Validation, and Methodological Justification</u>

Activities:

- Designed a controlled experimental plan to test the effectiveness of eco-friendly household treatments (heat, mild detergent, and vinegar) on reducing protein residues on animal fur
- Developed a complete draft of the procedure, materials list, variables, and preliminary data tables prior to experimentation
- Evaluated chemical safety considerations related to the Biuret assay, including reagent handling, personal protective equipment, and waste disposal
    - We had planned to use cooking pans in conducting our testing to minimize potential spillage on counter tops BUT apparently biuret should not be in contact with cooking pan metal (potential negative chemical reactions)
- Postponed experimental work to allow for discussion with my chemistry teacher and confirmation of safe, appropriate laboratory practices

Observation / Reflection:

- Although the plan was to begin testing during the Christmas break, reviewing safety documentation and recognizing gaps in our science lab tools at home plus our current understanding of chemical handling led us to pause experimentation
- Consulting an experienced chemistry teacher before proceeding will help ensure the project will be conducted safely, responsibly, and in alignment with laboratory standards
- Personal reflection: Choosing to delay experimentation actually strengthened the scientific integrity of our project and reinforced that responsible decision-making is an important part of being a scientist

Learning:

- Strong experimental design includes safety validation and expert consultation before data collection begins
- Preparing data tables and defining required measurements in advance improves precision, consistency, and reproducibility

- Scientific method includes knowing when not to proceed until uncertainties are resolved

December 26, 2025 –Methodological Decision: Use of Total Protein as a Proxy for Allergenic Proteins

Scientific Rationale:

- Specific pet allergens such as Fel d 1 (cats) and Can f 1 (dogs) are individual proteins that require specialized immunological assays such as an ELISA (Enzyme-Linked Immunosorbent Assay). This is not accessible in my high school or our home laboratory setting
- Accessibility Workaround: This project will instead measure total protein extracted from animal fur using the Biuret assay, which is a validated, quantitative, and education-appropriate colorimetric method
- Because allergenic proteins contribute to the total protein present on fur, reductions in total protein provide a scientifically justified proxy for assessing treatment effectiveness

Learning:

- Adapting methodology to real-world restrictions while keeping meaningful scientific comparisons is an important element of experimental science
- Identifying these methodological limitations strengthens credibility and demonstrates solid scientific reasoning

Next Steps:

- Set up reminder on procedure floor to prepare a protein standard curve using Bovine Serum Albumin (BSA). If BSA is not possible then collagen, gelatin or egg white albumin would be more accessible reference standards
- Apply the Biuret assay consistently across all treatments and fur types using controlled volumes and conditions
- Analyze treatment effectiveness using quantitative comparisons and document the scope and limitations of our findings

January 5–8, 2026 – Safety, Tools, and Method Adjustments

Activities:

- printed out some technical research resources to discuss and highlight
- Reviewed our draft science project (materials and process) with my chemistry teacher (Ms. Medd) to ask to borrow some materials and also to better understand safe handling of Biuret assay. She recommended small volumes, gloves, goggles, ventilation, and avoiding food prep areas.
- Asked to borrow school scale for accurate fur measurements (our kitchen / food scale wasn't accurate enough)
- Read product descriptions and reviews on Amazon and realized that the available Amazon colorimeters that we thought we could use is not suitable for our testing;
    - Accessibility Update - We will instead use a free iPhone Color Meter app
- Determined that BSA (for creating protein curve) is not available locally in stores or Amazon.ca. Decided to use egg white for protein reference curve

- Discussed and rewrote a big portion of materials and procedure based on above

Observation / Reflection:

- Adjustments emphasized the importance of flexibility and creative problem-solving
- Personal connection: refining the project made it feel more achievable, safer, and practical
- Proud that the project can now be performed more safely by us and replicated safely by other students at home. Due to not being able to purchase some of the planned lab tools / solutions, we updated our procedure to be more accessible for other students

January 9, 2026 – Fur Collection and Key Observations

Activities:

- Brushed Bella and collected additional fur. Someone knocked at the door; Bella became startled and shed a large amount instantly. Cleaned fur from hands and shirt with 3M lint roller

Observation / Reflection:

- Came up with a potential future research idea: study stress-induced shedding (without purposely causing stress)
- Thought of another potential research idea: how fabrics attract or repel fur, which could help people reduce allergen exposure around animals
- Personal reflection: Bella is enjoying this science project. She loves the extra grooming and attention she is getting

Learning:

- Everyday events can inspire meaningful scientific questions
- Observing how our cats shed fur under different conditions (like Bella shedding suddenly when startled or the horse shedding less when it's cold outside) helped us design a more consistent experiment. These observations are teaching us which variables to control, how to collect samples more reliably, and even helped us brainstorm new questions for future science projects, such as how stress and weather affect shedding or how different fabrics attract fur

Next Steps:

- Organize fur by type and treatment
- Measure fur to ensure that we will have enough. We only received small bags of horse and dog fur.
- Practice measurements with Bella's fur
- Conduct controlled testing using Biuret all in one day to minimize required exposure

January 9, 2026 – Learning to Measure Protein Using ColorMeter App

Activities:

- Downloaded and explored the ColorMeter app to measure Biuret assay color intensity.
- Tested app functions: RGB values, hue, brightness, and how each correlates to protein concentration.

- Tested different backgrounds (white, black, neutral) and lighting conditions to find the most consistent setup.
- Practiced measuring color from egg white dilutions to simulate standard curve readings.

Observation / Reflection:

- RGB values varied slightly depending on background and light intensity; using a fixed white background and consistent phone angle minimized variability.
- The Biuret violet color is most reliably tracked using the blue channel, though recording all RGB channels allows cross-checking for errors.
- Using a simple box or tray with white paper helped standardize measurements, prevented glare, and kept samples contained.

Learning:

- Consistency in background, lighting, and phone angle is critical for reproducible color measurements.
- The blue channel is the best indicator for Biuret assay, but capturing all channels adds redundancy and supports troubleshooting.
- Standardized color measurements allow conversion of color intensity into protein concentration when combined with a standard curve from BSA or alternative dilutions.

Next Steps:

- Prepare serial dilutions of BSA alternative to calculate protein levels diff equivalents
- Measure each dilution with the ColorMeter app under standardized conditions.
- Record RGB/blue channel values for each dilution and plot color intensity (y-axis) vs. protein concentration (x-axis) to create a standard curve.
- Use the trendline equation to convert color readings from treated fur samples into protein concentrations.

Date: January 10, 2026

Experiment Focus: Selecting a consistent, accessible protein source to replace BSA

Activities:

- Reviewed three potential protein sources available at home: egg white, Organika collagen peptides, and Great Lakes unflavoured gelatin.
- Evaluated each source for accessibility, consistency, and suitability for protein assays in a high school setting.
- Compared chemical composition and expected behavior in colorimetric tests.

Observation:

- Egg white: protein present but variable concentration, foams easily, cloudy, and spoils quickly.
- Collagen peptides: dissolves easily but hydrolyzed, may give weaker or inconsistent color reactions.
- Gelatin: nearly pure protein, dissolves uniformly, chemically closer to intact proteins, easily standardized, commonly used as a BSA substitute in school experiments (Green & Hughes, 2018; Harlow & Lane, 1999).
- Reflection:

- Gelatin provides the most reliable and reproducible option for a standard curve.
- Egg whites and collagen peptides introduce variability or potential interference that could affect assay accuracy and judge evaluation.
- Choosing gelatin ensures both scientific rigor and practical feasibility for a high school lab environment (American Chemical Society, n.d.).

Learning:

- Understanding how protein composition and purity affect colorimetric assays.
- Recognized the importance of reproducibility and standardization in experimental design.
- Learned to evaluate alternative materials when standard lab reagents are not accessible.

Next Steps:

- Prepare a stock solution of Great Lakes unflavoured gelatin at a known concentration.
- Create a series of dilutions to serve as a protein standard curve for the Biuret test.
- Record initial colorimetric readings to verify consistency before beginning the full experiment.

Date: January 10, 2026

Activities:

- We watched YouTube videos showing university scientists working with Biuret tests and protein experiments.
- When we realized there were very few videos exactly matching our experiment, we expanded our research to include protein measurement in food items, which gave us more practical and understandable examples.
- We did a detailed review of our draft materials list and experimental procedure to make sure they clearly matched each other and to confirm that we are obtaining the right number and size of materials such as test tubes.
- We calculated how much fur and how many solutions we will need for each trial and in total.
- We updated our procedure so it more clearly connects to the materials list, including the exact number and size of test tubes needed.

Observations:

- We could not find any Biuret experiments using animal fur, but food protein testing videos helped us understand the process better.
- Our initial calculations indicate that we need at least 8 grams of fur per sample type. We don't have access to the good scale yet but estimate this is likely about 1/2 full med sized ziplock bags.
- We currently have enough fur for one sample type, Bella (domestic short-haired cat), but not enough for the other fur types.
- Regarding the heat method, our research sources suggest protein denaturation may require temperatures close to >=80°C, which seems high and potentially unsafe to maintain for a school experiment.

Reflection:

- Broadening our research is helping us design our procedure and testing techniques since exact examples don't seem to exist.
- Realizing the fur quantity issue now is important because using too little sample would affect our ability to complete our planned testing and / or the accuracy of our results.
- Safety is a priority, so we need to balance scientific accuracy with realistic school lab conditions.

Learning:

- We learned that protein testing methods are used in many fields, including food science, which can be adapted and applied to our experiment.
- We improved our skills in calculating material amounts and scaling procedures properly.
- We developed a better understanding of how heat affects protein structure and denaturation.

Next Steps:

- Groom our longhaired ragdoll cat more and contact additional pet owners to try to collect more fur samples or decide whether we need to reduce the number of fur sample types.
- Confirm which supplies we can borrow from our school lab and list exactly why each item is needed, including test tube size and quantity. We will purchase any other remaining items online.
- Review scientific sources to determine whether using 60°C instead of higher temperatures can still produce reliable results while keeping the experiment safe.
- Finalize our updated procedure and materials list before beginning testing.

<u>Jan 11 2026</u>

- We double-checked our calculations for required fur mass and realized there was an error. We actually only need 4 grams per fur type, not 8. I haven't borrowed the accurate scale from the school yet but think we are still low on dog and horse hair.
- Our project focus has evolved greatly over the past two months. It changed due to not having access to elaborate science testing equipment and also changed with our interest based on ongoing research. We discussed our project focus and realized that the current fur types dog, cat and horse may be too broad since horse hair isn't as common inside homes which is what our project is now focusing on. We are considering removing horse hair which may be a blessing anyway since our supply is too low. We might instead see if we can get another sample of dog hair to replace the horse hair. This will result in us having two cat samples and two dog samples so might be better scientifically. If we replace or remove the sample, we will need to also update our draft procedure and raw data input templates accordingly.
- We also spent time reviewing our report for spelling, grammar and inconsistencies. Since we made lots of updates over a period of time, there was a lot of cleanup required. We're not done yet.

- We also reviewed the current reference list for completeness and to remind ourselves of all of the resources we have. All our info resources have been obtained online or by talking to teachers. Through google I found some potentially helpful books to refer to but they are not available in my local library system.
- I also printed out our current procedure and materials list. I plan to bring it to school to review with my teachers again. I'll review the materials list with my chem teacher to see what materials I can borrow from the school over a weekend in late Jan to complete our testing phase. The remaining materials will be purchased online. I also plan to meet with a senior biology teacher (who will be teaching me Bio 30 next term) to review our preliminary background research and procedure and to ask for advice into different angles that I might be missing and should further research and also that our procedure is safe and solid scientifically. I likely won't be able to meet with them until Friday because I am very busy with music lessons and exams next week.
- Used AI to create a chart summary of different methods to test for protein in items. This resource is helpful to ensure that we're considering more angles and not missing anything in our research.
- I also watched some of the training videos on the CYSF website to understand how to use the online submission form and created a tips sheet as a reminder for us when we start building our online report.
- Brushed my cat Bella today. Had a pile of the brushed off fur samples and she turned around and quickly ate it. This created a new future animal science question . Why do animals eat gross things, like their own fur? Here she ate shed fur but other days, she tries to eat Christmas tree pieces or crinkly plastic or other weird things. Maybe that'd be a good future research science project. Will add that to my list.
- Reviewed materials listing in detail and calculated volumes. Note that I'll need more testing solutions that originally specified because there's 16 samples per treatment x 10mL. So I actually need about 200 mL of each.
- Requested more dog fur from our friend. She said she will give more dog hair and photos. His name is Bond and he is a Czech Shepard . He will be 5 years old in March . We received photos of him too for our sample page.
- Reviewed procedure and discussed focus. We decided we will remove the horse hair because it doesn't align well with where our project is now. We will update the materials and procedure section to reflect this update. Now there are 48 samples instead of 64. 4 reps x 4 treatments x 3 fur samples . Updated materials and procedure sections to reflect this change.
    - ***SUBSEQUENT UPDATE MADE - This count reflected a draft design; the final experimental run used three replicates per treatment as documented in the Jan 24 testing entry.***

Jan 12, 2026

Activities:

- Reviewed the planned 80 vs 60 °C water bath heat treatment and evaluated its effectiveness for protein denaturation based on background research.

- Reviewed online sources looking for information on steam / moist heat and lower-temperature water treatments.
- Reviewed McDonnell & Russell (1999), which explains why moist heat (steam) denatures proteins more effectively than warm water due to improved heat transfer and disruption of protein structure (https://doi.org/10.1128/CMR.12.1.147).
- Reviewed the Instant Pot Pro user manual, which confirms that the Steam function on High produces steam at approximately 100 °C under non-pressurized conditions (Instant Brands Inc., 2021).
- Compared a low-temperature water bath with steam in terms of protein denaturation efficiency, reproducibility, and household relevance.
- Revised the experimental procedure to replace the 60 °C water bath with a steam-based heat treatment using the Instant Pot Steam function set to High.
- Updated materials, variables, and safety considerations to reflect the new steam treatment method.
- Standardized post-treatment rinsing across all treatments and confirmed that no pre-rinsing would be performed.

Observation:

- Scientific literature indicates that moist heat causes more effective and irreversible protein denaturation than lower-temperature water baths.
- Steam provides higher and more uniform heat (~100 °C) than a 60 °C water bath and better reflects common household steam-cleaning practices.
- Elevating fur samples above water allows exposure to steam without uncontrolled protein loss from soaking.
- Clearly specifying the Steam-High setting improves repeatability and reduces ambiguity.
- Reflection:
- Supporting the procedural change with both peer-reviewed literature and manufacturer documentation strengthens the scientific justification and credibility of the experiment.
- Using steam improves biological relevance and increases the likelihood of observing a clear, measurable reduction in extractable protein.
- This refinement reduces the risk of inconclusive heat results while maintaining safety and ethical standards.

Learning:

- Learned that the effectiveness of heat depends on both temperature and method of heat transfer, not temperature alone.
- Gained experience using scientific literature and manufacturer manuals to support experimental design decisions.
- Improved my ability to justify procedural changes using mechanistic biochemical reasoning.
- Next Steps:
- Incorporate McDonnell & Russell (1999) and the Instant Pot Pro user manual into the background research section and reference list.
- Update the revised procedure and materials list in the project plan.

- Conduct a short trial run of the steam setup with my mom to confirm timing, safety, and consistency.
- Print the revised materials and procedure list to discuss with my chemistry and biology teachers when requesting equipment and supplies.

Jan 13, 2026

- I created a pet fur sample summary page to show where we sourced pet fur from with their cute pictures and bios
- Self-reviewed research section noting it was very long and a bit repetitive. Significantly shorted research section so it will fit on board better. Printed the long version also so we can reference back to it if needed or if we decide we need more detail.

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Jan 16, 2026

- Met with my Chem teacher at school today. Reviewed materials list together. She confirmed that I can borrow all of the required equipment related supplies. I plan to borrow the equipment on Friday Jan 23 to complete the testing that same weekend.
- I still need to meet with my biology teacher to review my draft procedure and discuss biuret reagent safety. Specifically, I am concerned about how I can safely clean the borrowed equipment that has been in contact with the reagent and whether that diluted run off (if water washing) is considered a waste product that needs to go in a waste container for proper disposal or can go down the kitchen drain.
- I also realized that I should further edit the procedure to make it more clear. Right now, it's long and a bit confusing if someone else was going to follow. I will self review it again this weekend before I meet with my bio teacher.
- My mom also shared an interesting metaphor with us about this science experiment that I think might be helpful for explaining the process to other people, especially younger students
    - Dirty clothes metaphor (the dirt on the clothes represents allergens / proteins on the pet for)
    - Imagine you have muddy clothes. When you wash the clothes with soap, the muddy water goes down the drain.
    - To see how clean the clothes really are after you wash them, you could put them into a clean bucket of water and gently swirl them.
        - If the water continues to get muddy, the clothes still have a lot of dirt on them.
        - If the water stays clear, it confirms most of the dirt was already washed away.
    - For our experiment, the fur is like the clothes and the protein is like the mud. The final clean bucket of water is the extraction step that shows how much mud / protein was still stuck on the clothes / fur after washing.

Jan 18, 2026

- Reviewed and printed cited reference materials for binder. This took a LONG time.
- Reviewed procedure and clarified wording around how the samples will be tested (on batches for efficacy and to minimize confusion / risk of manual error)
- Also reviewed and clarified variables section
    - Independent variables: Treatment type (distilled water, vinegar, detergent, steam heat); fur type
    - Dependent variable: Extractable protein concentration (estimated via Biuret assay color intensity)
    - Controlled variables: Sample mass, treatment time, extraction volume and time, Biuret reagent ratio, lighting conditions, and measurement method

Jan 20, 2026

- Received additional dog fur sample from our family friend (same dog as before). They used a furminator to brush him this past weekend. They said he usually sheds a lot but not as much this time of year and that this is primarily his undercoat. I wonder if time of year could impact the amount of allergen / dander in the fur. Future science experiment ideas or tips for future students conducting this experiment - collect for samples in spring when animals are shedding more.

Jan 21, 2026

- Met with Mr. Verhees (he will be my bio 30 teacher next semester) to review science project safety requirements. He shared a lot of info on biuret reagents and walked me through safety standards.

Jan 22, 2026

- Reviewed materials listing again compared to procedure to ensure I know total amounts of solutions and completeness of equipment needed.
- We are doing the testing Saturday and borrowing supplies from my chem teacher tomorrow (Friday).
- The Biuret reagent we ordered on Amazon is late so we're going to borrow from the school and then give them our bottle once received. My school has been very supportive.
- We were originally going to use a plastic water bottle for our biuret waste. My dad instead gave us a very large sturdy wide mouthed glass sauerkraut jar that has a lid and will be much easier / safer for us to pour waste into and transport it back to school for proper disposal.

Jan 23, 2026

- Met with my Chemistry teacher (Ms. Medd) at lunch to box up all of the supplies we are borrowing. My dad picked me up from school because the box was too big to reasonably fit on the school bus.

- The big bottle of Biuret we ordered from Amazon was delivered today so we won't have to borrow from the school after all. Read safety instructions cautions on back of bottle.

Jan 24, 2026 - Sat - It's Finally Testing Day!!!

- Activity: Full Experimental Testing Day - Protein Extraction & Biuret Assay
- Time Spent: ~8.5 hours active testing + ~2.5 hours setup/cleanup
- Testing Day Overview
    - Today was our primary experimental testing day. Our goal was to apply different treatments (distilled water, vinegar, detergent, and steam heat) to fur samples, extract soluble protein from the treated fur, and estimate relative protein levels using the Biuret assay and smartphone colorimetry.
    - We methodically followed our procedures as written with a few minor deviations as noted below. All procedural steps, deviations, and observations have been documented in sufficient detail to allow another student to replicate this experiment using only this logbook.
- Pre-Testing Preparation
    - Before starting, we rewatched several YouTube videos on how the Biuret protein test works to refresh our understanding of what color changes to expect and how strong the reactions should look. This helped us feel more confident about interpreting results during testing.
    - We reread our written procedure and printed two copies so both of us could easily follow along. We wrote our names beside each step so it was clear who was responsible for each part of the process.
        - Kiana - Sample Treatment Station - primarily responsible for setting up samples (distilled water, vinegar, etc)
        - Mom - Instant Pot Station - primarily responsible for using the Instant Pot (Steam Heat) and using the managing the camera (colormetric app & taking pics / video)
        - Katie - Biuret Testing Station - primarily responsible for the biuret reagent reactions and recording results into the data templates
        - Note - Although we helped each other during physically demanding or time-sensitive steps, Katie was the only person who handled the Biuret reagent and Biuret waste to maintain safety and consistency.
    - We also printed all of our data table templates ahead of time so we could record results quickly without stopping to reorganize.
- Workspace Setup
    - We set up three separate workstations in the kitchen to stay organized and prevent cross-contamination:
        - 1 - Biuret testing & photo station
            - White plastic layered background
            - Smartphone with color analysis app
            - Test tubes and Biuret reagent
        - 2 - Sample treatment station

                - Fur samples & cups
                - Beakers, stirring rods, treatment solutions
                - Deviation from plan: We originally planned to use glass beakers for all treatment steps. During testing, we switched to disposable cups at the treatment station so we could prepare multiple samples at once instead of waiting for beakers to be washed and dried. All cups were clearly labeled to prevent mix-ups.
            - 3 - Instant Pot & Steam heat treatments
                - Rinsing and cooling area
- We organized all supplies from home and the equipment borrowed from school before starting. We took some photos of the setup before starting.
- Unexpected Problem & Fix (Pipettes)
    - During setup, we realized we had forgotten the rubber bulbs needed for fine pipette measurements. Because it was Saturday, the school was closed, which could have stopped the experiment. Instead of stopping:
        - We called several local pharmacies
        - Found one that sold pipettes
        - Purchased replacement pipettes immediately
        - Also bought measured droppers as backups.
    - Lesson learned: Always double-check the materials list multiple times before testing day, especially for small but critical items.
- Gelatin Calibration Curve – Modified the procedure
    - We prepared the gelatin calibration curve first.
    - Applied the Biuret assay to fur sample extracts prepared from different treatments (water, vinegar, detergent, heat).
    - Measured color intensity using ColorMeter app under standardized white background and lighting.
    - In subsequent analysis, we will convert blue channel color readings into protein concentrations using the gelatin standard curve.
    - Originally, our plan used **1 g of gelatin** in 100 mL of water, but when we tested this concentration with Biuret reagent, the color change was very weak. There was little visible difference between concentrations using the phone app. To make sure the problem was not the Biuret reagent itself, we tested a stronger gelatin mixture with Biuret, and it turned very purple, confirming the reagent was working. We kept the purple test tube for reference, which you can see in some of our testing photos.
    - Because the colour change on the 1g / 100 ml of water curve looked weak, we changed the gelatin stock concentration to 3 g in 100 mL of distilled water to increase the signal strength. This worked much better for color detection. We made a note on our data sheet of the change so that we could recalculate the concentrations for the curve later.
    - Challenges with Gelatin
        - Took longer to dissolve

        - Slightly bubbly, making precise volume measurements harder
        - Difficult to see small volumes in test tubes
        - Gelatin tended to stick to the bottom of containers
        - The solution thickened over time between early and late curves (realized during later analysis)
    - We used the same Biuret ratio (0.20 mL sample + 1.80 mL reagent) for both calibration standards and fur extracts to ensure consistency.
    - Quality Control & Standardization
        - Same white background used for all color readings
        - Same phone, app, and lighting conditions used throughout
        - Triplicate blue-channel readings taken for each tube
        - Reagent blank prepared (distilled water + Biuret)
        - Strong gelatin-Biuret tube kept as a visual reference
    - Learning:
        - Using a standard curve allows raw color readings to become meaningful, quantitative protein measurements.
        - Even simple home-based methods can generate powerful data when calibration and careful handling are applied.
        - Documenting all steps, observations, and reflections strengthens the scientific rigor of the project.
        - Biuret does not measure very low protein concentrations well
        - Gelatin is not ideal for precision work. BSA (preferred but unavailable); gelatin was therefore used consistently as the standard. BSA was not available to us to borrow from school or purchase on amazon but science teachers might be able to order it in the future from a special supplier.
- Sample Treatments – What We Observed
    - We processed samples in organized batches by fur type to maintain consistent handling and minimize cross-contamination. Within each fur type, individual samples were randomly assigned to treatment groups before testing. Treatments were then applied in a fixed sequence within each batch to maintain consistent timing, temperature, and reagent conditions across replicates. Reusable glassware was rinsed between usage.
- Deviation – Rinsing
    - Planned: Two rinses
    - Actual: One rinse
    - Reason: Time constraints and limited distilled water
    - Applied consistently to all samples
- Deviation – Replicates
    - Planned: Four replicates per treatment
    - Actual: Three replicates
    - Reason: Dog fur sample (from a family friend) was too small to support four full replicates

    - Note - Final experimental design used three replicates per treatment for all fur types; earlier plans for four replicates were revised prior to testing due to sample availability.
- Detergent Treatment Observations
    - Dog fur became extremely sudsy and was difficult to rinse
    - Short-haired cat fur was coarser and absorbed less soap
    - Detergent solution became slightly yellowish, suggesting removal of dirt or residue
    - Visually, detergent appeared to clean fur effectively
- Steam Heat Observations
    - Cat fur clumped together due to thickness but steamed evenly
    - Dog fur was finer and fluffier
    - Opening the Instant Pot after steaming dog fur produced a strong odor, suggesting a stronger thermal effect
- Protein Extraction & Biuret Testing
    - After treatments:
        - We extracted protein by stirring samples in distilled water for 10 minutes
        - Transferred clear extracts to test tubes
        - Mixed 0.20 mL extract + 1.80 mL Biuret reagent
        - Prepared a reagent blank
    - We measured color using the ColorMeter app and recorded triplicate blue-channel values.
    - Visually, most samples looked very similar. Differences were only detectable using the phone app.
    - Lesson learned: Smartphone colorimetry is essential; visual inspection alone is not reliable.
- Data Recording Issue
    - One value was initially recorded in the wrong row. We caught this quickly and corrected it. After that, we verbally confirmed the sample ID out loud before recording each measurement.
- End-of-Day Reflection
    - Total time today: ~11 hours
    - Organization and teamwork were essential
    - Flexibility was required when problems occurred
    - Writing deviations immediately helped maintain scientific honesty
- Our parents took us out for a very late supper to celebrate

<u>Jan 25, 2026</u>

- Date: January 25, 2026 (Sunday)
- Activity: Data Entry, Analysis, and Interpretation
- Data Entry & Verification

    - We entered all results into Excel and generated the calibration curve. Initially, the curve looked skewed. After checking the raw data, we discovered that replicate 3 values had been entered in reverse order.
    - We corrected the data entry and regenerated the curve. After correction, the curve appeared strong and consistent.
    - Lesson learned: Data checking is just as important as lab work.
- Analysis & Detection Limit Issue
    - This project does not directly measure specific allergens (e.g., Fel d 1), but rather evaluates relative changes in total extractable protein as a justified proxy within the accessible laboratory limitations.
    - During calculations, we realized that most fur extract samples were below the reliable detection range of the Biuret assay.
    - Although this was disappointing, it revealed an important limitation of the method. At the start of the experiment, protein levels were unknown, so we used a standard Biuret ratio. In hindsight, this experiment functioned as a pilot study to assess assay sensitivity.
    - Low readings do not mean protein was absent; they indicate that the method was not optimized for very small protein amounts.
    - Because many values were below the assay's reliable detection range, results are interpreted qualitatively and comparatively rather than as absolute concentrations.
    - Replicate measurements showed some variability, particularly near the detection limit, reinforcing the need for higher assay sensitivity in future work.
- Controls & Method Validity
    - Distilled-water-treated fur samples served as controls
    - A Biuret reagent blank established the zero-protein baseline
    - Strong gelatin-Biuret reaction confirmed reagent functionality
    - Therefore, low readings were due to method sensitivity, not equipment failure.
- Limitations of Conclusions
    - Because many values were below the assay's detection range:
        - We cannot report precise absolute protein concentrations
        - Conclusions focus on relative trends and percent reduction, not exact values
        - Gelatin is not an ideal calibration standard for fur proteins
- Despite these limitations, trends across treatments were consistent with expectations.
    - **If validated with higher-sensitivity methods, these findings could result in safer, eco-friendly approaches to reducing allergen exposure in homes without harming animals.**
- Sources of Error & Limitations
    - Gelatin thickening and sticking affected calibration consistency
    - Biuret assay limited sensitivity at low protein levels

    - Reduced rinsing may have left detergent residue
    - Smartphone color measurements subject to small lighting/angle changes
    - Human fatigue and data-entry error during long testing sessions
- Improvements & Redesigned Next Iteration
    - If we repeated this experiment:
        - Use a purified protein standard (BSA or egg white protein)
        - Increase extraction concentration or sample mass
        - Adjust Biuret ratios or use a more sensitive assay
        - Prepare fresh calibration standards throughout testing
        - Add extra rinse for detergent samples
        - Split testing across multiple days to reduce fatigue
- Future Research Purpose & Applications
    - Based on the results and limitations identified in this study, the future purpose of this research would be to optimize protein detection sensitivity and directly relate treatment effectiveness to real-world allergen reduction.
    - Future experiments would focus on increasing assay sensitivity through sample concentration, alternative protein assays, or purified standards, and on expanding applications beyond fur to pet bedding, blankets, and grooming tools. Because the treatments tested are eco-friendly and non-toxic, this research has practical relevance for reducing allergen exposure in homes while maintaining animal safety.
    - Longer-term research could investigate seasonal shedding effects, stress-related shedding, and how different fabrics retain or release allergenic proteins, helping allergy sufferers make informed decisions about pet care and home environments.
    - Future project ideas could include
        - using these treatments for cleaning pets beds etc because they're non toxic safe for animals and would reduce allergens on their bed blankets etc
        - Understanding these treatments more if they're effective will empower more people to safely clean their pet areas, homes of dander with less chemicals and potentially safely clean it at the source on the animal during grooming
        - Designing an instant pot type steamer to clean pet beds and toys

Overall Reflection (Nov–Feb)

- All methods and deviations are documented so the experiment can be reproduced without additional instruction.
- This study evaluates protein removal under controlled conditions and does not measure allergen exposure in living environments.
- We encountered many roadblocks, most resulting in this project being easy to reproduce with easy to access supplies.

- We narrowed range, removed UV light, focused on common household treatments, and targeted allergens more relevant to homes. This evolution strengthened the project's clarity and feasibility.
- Additionally, the challenges improved our scientific thinking. Our project evolved as we learned more and adapted to realistic constraints.

Feb 8, 2026 - Science Fair Event Prep - Ideas & Reminders

Youtube Videos - Reminder to watch these again. These videos were used for personal understanding and method verification and are not cited as primary scientific sources. They're not used in our final report so we don't reference them but they're helpful as a refresher for when we need to discuss the biuret reaction steps / process with judges and others at the science fair.

- Curtin University. (n.d.). EXERCISE 5, Activity 4: BIURET TEST: Presence of Proteins [Video]. YouTube. https://youtu.be/p9Mq681i_-4

Used for:Demonstrates the Biuret test procedure, showing colour changes that indicate protein presence. Reinforces understanding of assay principles and visual interpretation.

- Protein Quantitation via the Biuret Assay | Biochemistry. (2024, May 22) [Video]. YouTube. https://youtu.be/PjLxg8Lul3U

Used for:Provides a detailed walkthrough of the Biuret assay, explaining chemical principles and step-by-step procedure. Useful for visual learners and method validation.

- Smith, J. (2024). Protein quantitation via the Biuret assay [Video]. YouTube. https://youtu.be/PjLxg8Lul3U

Summary: Explains how to quantify protein concentration using the Biuret assay, including calibration curve preparation. Supports the method used in this project.

- Zaitsev, I. V. (2024, July 25). EXERCISE 5, Activity 4: BIURET TEST: Presence of Proteins [Video]. YouTube. https://www.youtube.com/watch?v=jzxm803GEY8

Used for:Shows the Biuret test in action, confirming experimental steps and expected colour changes. Helpful for comparing results during the project.

Trifold Board

- Finish printing off our board resources to paste onto our board. We are planning to use white, blue and purple to symbolize the biuret reaction.

Prepare Biuret Test Tube Visuals

- The CYSF Safety Committee confirmed that we can bring small vials of the biuret reaction to the science fair as long as they're sealed and behind glass. This will be a fun demonstration to show judges and other students. They can visually see the colour difference plus try the Colormeter app on our phone to understand how the colour reader works.

Fun Science Fact Cards - Protein, Denaturation, & Biuret

- For our booth, we can prepare fun fact cards with questions on one side and the answers on the other side. People can quiz themselves.

Logbook

- Will print this logbook to include in our binder.

What to Bring - print this

- Trifold

- Logbook binder
- Biuret sample display box
- Fun Science Fact Cards
- Water bottles / Lunch
- Phones

Our Logbook Conclusion

- This logbook documents the full development of our project, including idea formation, background research, ethical considerations, experimental planning, revisions, and observations. Throughout the process, we faced real challenges related to safety, equipment access, and feasibility. By consulting teachers, reflecting on our options, and revising our methods, we were able to adapt our project responsibly.
- Observations of our cats, differences in fur type, and stress-related shedding informed how we controlled variables and planned experiments. Adjusting our methods (from UV light to heat, BSA to egg white, and colorimeter to phone app) demonstrates resilience, problem-solving, and our creative scientific thinking.
- This project showcases how personal experience and curiosity can drive meaningful science, highlights the importance of ethical and reproducible research, and provides insights that may help allergy sufferers safely enjoy pets.
- Key Takeaways:
    - Flexibility and adaptation are essential to problem-solving in science
    - Personal curiosity and lived experience can inspire impactful scientific inquiry
    - Project evolution demonstrates resilience, creativity, and critical thinking
    - Ethical interactions with animals reinforce that science can be humane and personally meaningful
    - Observations and methodological adjustments connect directly to reproducibility and experimental rigor
    - The project encourages future research, including the impact of stress, fur type, and fabric interaction on allergen exposure