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JOINT RESEARCH PROGRAMME 2021/2022

2022 Project Final Report

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Abstract

Waste products can be reused as secondary products (food additives) if an efficient preservation method is available and economically feasible. The project aimed to preserve food and reduce waste by reusing agricultural by-products through dehydration and purification. The project assessed the suitability of different drying processes for preservation in terms of food quality and cost. Site visits were conducted during this project to understand the problem better and identify opportunities. After site visits, samples (cow bone broths and egg whites) were collected for studies. They were dried using freeze- and spray-drying methods and then sent for shelf life, protein, and moisture content studies. In shelf-life studies, spray-dried samples demonstrated TVC (total viable count) growth, whereas freeze-dried bone broth samples and egg white samples demonstrated better TVC growth, yeast, and mould count. Spray-dried samples spoil faster, whereas freeze-dried samples destroy all microorganisms. Amino acid distribution is good in both spray and freeze-drying. Spray drying denatures complex proteins. From protein and amino acids analysis, the study results indicated no significant amino acid differences between the spray egg whites and the broth samples when compared to each other. However, there was a clear difference in protein percentage between the two egg whites, with the freeze-drying egg whites having a higher protein percentage of 80.6%, likely due to the higher temperatures. In contrast, the bone broth had a lower protein percentage due to the overnight boiling of the egg whites.
Results indicate that freeze-drying is a better option for extending shelf life and retaining nutrients. However, the energy requirements for this application are still to be determined. The significance of the project was to contribute towards reaching sustainable development goals, specifically goal number 2 (end hunger, achieve food security and improved nutrition and promote sustainable agriculture) and goal number 12 (Ensure sustainable consumption and production patterns).

1. Introduction

Food waste plays a pivotal role in the world hunger crisis, as roughly 1.3 billion tons (30% of produced food worldwide) of food are wasted in the food chain every year. Waste products from agriculture (agro-processing) can be used in secondary products as food additives or additives if an efficient preservation method is available and economically feasible. On this project, CPUT researchers and students collaborated with Farmer Angus, a livestock and poultry farmer in Stellenbosch, Western Cape. The opportunity for waste reduction was introduced by Farmer Angus, related to a waste of cow bones and egg whites produced on his farm. Although some processes have been introduced to create broths from cow bones, the jelly-like broth is subject to spoilage due to its products' high moisture content and short shelf life. Similarly, egg whites produced during the separation of egg yolks are produced in large quantities and are prone to spoilage.

Farmer Angus generates large quantities of waste in the food chain and ultimately incurs a financial loss. Farmers could benefit significantly from food processing, which prolongs agro-products shelf-life. This project envisioned to (i) determine the types, volumes, and seasonality of agricultural animal wastes from small and medium farms in the Western Cape using a combination of desktop studies and site visits; (ii) compare the effect of spray-drying and freeze-drying techniques on moisture content, particle size distribution and protein structural integrity of egg whites and bone broths. Furthermore, the project envisioned to address 2 of the United Nations SDGs. Firstly, to end hunger, achieve food security, improve nutrition, and promote sustainable agriculture (SDG 2). Secondly, to ensure sustainable consumption and production patterns (SDG 12).

2. Aim

To valorise and improve the shelf-life of livestock by-products.

3. Research questions and objectives

Question 1: How much animal waste is produced by farmers (livestock and poultry) in the Western Cape?

Question 2: Which drying method is better in terms of protein quality, concentration, and energy consumption?

Question 3: How does the moisture content of dried broth and egg white affect shelf life?

Question 4: Will PSD affect the solubility of a mixture of collagen, broth, and egg whites?

4. Research approach and methods

Fresh eggs, on the same day of laying, were collected from Angus farmer in Cape Town, Western Cape, South Africa, and all the chemicals used were of analytical grade.

4.1. Dehydration of egg whites by spray drying

A single-stage, tall-form co-current lab scale drier (Spray Mate, JISL) was used for spray drying. The atomisation was done with compressed air at a pressure of 24 psi with a dual fluid nozzle atomiser. The inlet air temperature was controlled by heating ambient air directly in an electrical heater. The inlet (120 °C) and outlet (80 °C) air temperatures were maintained during drying. The procedure was started with distilled water being fed into the system, and the outlet temperatures were adjusted by altering the liquid feed and atomising gas flow rate. After reaching the necessary outlet temperature, the feed solution was delivered into the spray drying chamber. The exit temperature was well controlled by adjusting the liquid supply flow rate (30ml/min).

4.2. Dehydration of egg whites by freeze-drying

The freeze-drying process followed the method described by (Karthik & Anandharamakrishnan, 2013). An ultra-low deep freezer (So-Low Environmental Equipment Co. Inc., Ohio) was used to freeze the egg whites for around 2 hours. The frozen sample was placed in a freeze drier (Heto Dry Winner) at 24 °C. The encapsulated powder was packaged in vacuum-sealed bags and stored at refrigerator temperature (41 °C).

4.3. Amino acids analysis

The amino acids were analysed using the method of Jajic et al. (2013) using high-performance liquid chromatography (HPLC). The mobile phase A was acetonitrile: methanol: water (45:45:10, vol. %); The mobile phase B consisted of 5.5 g of Na_2HPO_4 per 1 l of distilled water, adjusted to the pH 7.8 using 10 M NaOH solution.

4.4. Particle size distribution and size reduction

Different fractions of dried products were identified using a sieve of the following sizes: 6700□m, 2000 □m, 1400 □m, 1000□m, 600 □m and 425 □m. Size fractions above 425 □m were ground using a 200W coffee grinder until >95% of the dried sample passed a 425 □m sieve.

4.5. Solubility of dried products

A mixer at 100 rpm was inserted in a 500 ml beaker filled with 250 ml of deionised water. A sample size of 5g of spray-dried egg whites, freeze-dried egg whites, spray-dried broth and freeze-dried broth was added to the water. The time taken to dissolve the products was recorded.

5. Results

5.1. Shelf life

Shelf-life studies over a period of five weeks were carried out at the Agrifood Technology Station, Faculty of Applied Sciences, Cape Peninsula University of Technology, on product samples of liquid eggs, liquid broths, freeze-dried egg white and bone broths, spray-dried egg white and bone broths, and the results were as follows.

5.1.1. Microbial analysis on egg white and bone broth liquid samples

Table 1: Results of Microbial analysis on Egg White liquid as supplied for analysis on 31st of July 2023

Sampling and Testing Dates	No.	Sample	Average number of colony forming units (CFU) per gram		
			TVC (average)	Yeast (average)	Mould (average)
01/08/2023	1	Egg White Liquid	TNTC	10	0
15/08/2023	2	Egg White Liquid	TNTC	230	0
21/08/2023	3	Egg White Liquid	TNTC	465 000	0
28/08/2023	4	Egg White Liquid	TNTC	TNTC	0

Egg White Liquid samples tested over the five weeks showed unsatisfactory results for the Total Viable Count (TVC) from Day 1 of testing to Day 4 of testing as the TVC was too numerous to be counted (TNTC) see Table 1. Yeast counts for samples tested on Day 1 and Day 2 of testing showed satisfactory results, with values of 10 and 230 being within an acceptable range. However, from day 3, the values increased beyond the acceptable range and on day four, it was too numerous to be counted (TNTC) (see Table 1). But as the days went by, the number of yeast cells in the samples grew and multiplied, leading to higher counts of colony-forming units on samples tested on Day 3 and Day 4. Hence, unsatisfactory results. Mould counts showed promising results with no growth and zero counts from Day 1 to Day 4 of sample testing. Furthermore, the Bone Broth Liquid samples tested over the five weeks showed satisfactory results for the TVC, Yeast, and Mould counts. Although there was some presence and growth of TVC and moulds on the sample tested from Day 1 to Day 4, the number of colonies forming units was within acceptable levels.

5.1.2. Microbial analysis on freeze-dried bone broth and freeze-dried egg white samples

Table 2: Results of Microbial analysis on Freeze Dried Bone Broth as supplied for analysis on 31st of July 2023

Sampling and Testing Dates	No.	Sample	Average number of colony forming units (CFU) per gram		
			TVC (average)	Yeast (average)	Mould (average)
01/08/2023	1	Freeze Dried Bone Broth	0	0	0
15/08/2023	2	Freeze Dried Bone Broth	0	0	0
21/08/2023	3	Freeze Dried Bone Broth	0	0	0
28/08/2023	4	Freeze Dried Bone Broth	0	0	0

Table 3: Results of Microbial analysis on Freeze Dried Egg Whites as supplied for analysis on 31st of July 2023

Sampling and Testing Dates	Sample No.	Sample	Average number of colony forming units (CFU) per gram		
			TVC (average)	Yeast (average)	Mould (average)
01/08/2023	1	Freeze Dried Egg White	0	0	0
15/08/2023	2	Freeze Dried Egg White	0	0	0
21/08/2023	3	Freeze Dried Egg White	0	0	0
28/08/2023	4	Freeze Dried Egg White	0	0	0

Freeze Dried Bone Broth and Egg White samples tested over the five-week period from Day 1 to Day 4 showed outstanding results for the TVC, yeast and mould counts as the samples showed no microorganism growth (see Table 2 and Table 3). This implies the freeze-drying process efficiently killed all microorganisms present in the freeze-dried samples for this shelf-life study.

5.1.3. Microbial analysis on spray-dried bone broth and spray-dried egg white samples

Table 4: Results of Microbial analysis on Spray Dried Bone Broth as supplied for analysis on 31st of July 2023

Sampling and Testing Dates	No.	Sample	Average number of colony forming units (CFU) per gram		
			TVC (average)	Yeast (average)	Mould (average)
01/08/2023	1	Spray Dried Bone Broth	80	0	0
15/08/2023	2	Spray Dried Bone Broth	200	0	0
21/08/2023	3	Spray Dried Bone Broth	880	0	0
28/08/2023	4	Spray Dried Bone Broth	1460	0	0

Table 5: Results of Microbial analysis on Spray Dried Egg Whites as supplied for analysis on 31st of July 2023

Sampling and Testing Dates	No.	Sample	Average number of colony forming units (CFU) per gram		
			TVC (average)	Yeast (average)	Mould (average)
01/08/2023	1	Spray Dried Egg White	525	0	0
15/08/2023	2	Spray Dried Egg White	1000	0	0
21/08/2023	3	Spray Dried Egg White	2750	0	0
28/08/2023	4	Spray Dried Egg White	3630	0	0

TVC analysis done on the Spray Dried Bone Broth and Spray Dried Egg White samples showed some presence and growth of colonies. However, the number of colonies forming units was within acceptable levels (see Table 4 and Table 5). Furthermore, for the Yeast and Mould counts, there was no growth detected.

5.2. Proteins and Amino acids

Table 6: Dietary analysis

TEST TYPE	Wet egg white	Freeze dried egg white	Spray dried egg white	Bone broth	Freeze dried bone broth	Spray dried bone broth
Moisture (g/100g)	87.4	9.0	11.2	94.5	11.5	ND
Ash (g/100g)	0.74	5.45	6.60	0.38	6.54	ND
Total Fat (g/100g)	0.70	1.15	2.32	<0.19	1.23	ND
Saturated Fat (g/100g)	0.64	1.00	2.15	0.00	1.12	ND
Mono-Unsaturated Fat (g/100g)	0.05	0.11	0.16	0.00	0.10	ND
Poly-Unsaturated Fat (g/100g)	0.01	0.04	0.01	0.00	0.01	ND
Trans Fat (g/100g)	0.00	0.00	0.05	0.00	0.00	ND
Omega 3 Fatty Acids (g/100g)	0.000	0.000	0.000	0.000	0.000	ND
ALA (g/100g)	0.000	0.000	0.000	0.000	0.000	ND
EPA (g/100g)	0.000	0.000	0.000	0.000	0.000	ND
DHA (g/100g)	0.000	0.000	0.000	0.000	0.000	ND
DPA (g/100g)	0.000	0.000	0.000	0.000	0.000	ND
Omega 6 Fatty acids (g/100g)	<0.2	<0.2	<0.2	<0.2	<0.2	ND
Nitrogen (g/100g)	1.78	12.90	11.15	0.69	11.12	11.13
Protein (g/100g)	11.1	80.6	69.7	4.3	69.5	69.5
Total Dietary Fibre (g/100g)	<0.2	<0.2	ND	<0.2	<0.2	ND
Total Sugar (g/100g)	0.4	0.4	ND	0.5	6.1	ND
Fructose (g/100g)	<0.1	<0.1	ND	0.1	0.9	ND
Glucose (g/100g)	0.3	0.3	ND	0.1	0.3	ND
Sucrose (g/100g)	<0.1	<0.1	ND	0.2	4.8	ND
Maltose (g/100g)	<0.1	<0.1	ND	<0.1	0.2	ND
Lactose (g/100g)	<0.1	<0.1	ND	<0.1	<0.1	ND
Galactose (g/100g)	<0.1	<0.1	ND	<0.1	<0.1	ND
Trehalose (g/100g)	<0.1	<0.1	ND	<0.1	<0.1	ND

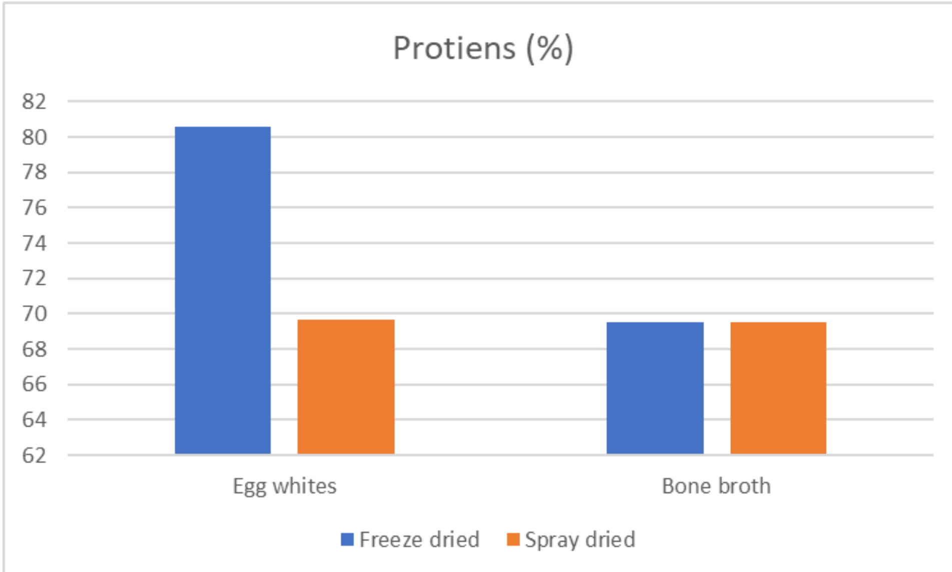

Figure 1: Freeze and Spray dried protein %

There were no significant differences in amino acids between the spray and freeze-dried egg whites and broth samples (Table 2). Both samples have a similar distribution of amino acids, and none of the drying methods seems to have a destructive effect on amino acids. However, a clear distinction exists in % proteins of egg white in favour of freeze drying over spray drying (Figure 1). Freeze-dried proteins (80.6%) were higher than spray-dried proteins (69.7%), likely due to the use of temperatures ranging between 55-70 ˚C, which would likely denature proteins. No such trend exists in bone broth because its initial preparation involved boiling overnight, leading to the denaturing of most complex proteins.

Table 7: Mixtures of dried egg whites, dried broth and collagen samples

	His	Arg	Ser	Gly	Asp	Glu	Thr	Ala	Pro	Lys	Tyr	Met	Val	ILe	Leu	Phe
Name	% g/100g	% g/100g	% g/100g	% g/100g	% g/100g	% g/100g	% g/100g	% g/100g	% g/100g	% g/100g	% g/100g	% g/100g	% g/100g	% g/100g	% g/100g	% g/100g
Collagen	0.47	3.28	0.96	10.42	2.92	4.34	0.67	4.32	8.84	1.81	0.29	0.44	0.98	0.82	1.54	0.84
Freeze dried egg white	0.55	1.71	1.99	1.17	4.22	4.37	1.37	2.21	1.81	2.69	1.16	2.00	2.38	1.78	3.22	2.01
Spray dried egg white	0.54	1.71	2.19	1.43	4.34	4.95	1.62	2.42	1.86	3.15	1.38	2.25	2.63	1.97	3.50	1.99
Freeze dried broth	0.34	2.06	0.82	6.68	2.00	3.77	0.59	2.79	5.34	1.16	0.32	0.47	0.83	0.52	1.31	0.76
Spray dried broth	0.32	2.26	0.81	6.56	2.47	4.17	0.81	3.05	4.91	1.22	0.26	0.44	1.06	0.61	1.56	0.89
Egg white (wet)	0.67	1.88	1.90	1.36	3.67	4.12	1.30	2.19	2.10	2.58	1.17	2.44	2.54	1.97	3.77	2.30
Broth (wet)	0.36	1.82	0.53	5.92	2.04	3.09	0.56	2.41	5.54	0.97	0.29	0.33	0.82	0.54	1.44	0.84

5.3. PSD, Solubility, and Energy requirements

Particle size distribution (PSD) is an important factor in selecting the most efficient method of drying. PSD will affect energy requirements, as dried products may require milling to achieve uniformity for mixing and solubility/ dissolution.. The PSD of spray-dried products (98.2% < 0.425mm) generally resembles collagen 94% < 0.425mm), while freeze-dried samples contain a wider range of PSD (8.2%-33.3% < 0.425mm). Freeze drying will require an additional 56.6 kWh/kg and 13.3 kWh/kg for bone broth and egg whites respectively to mill dried products to a PSD like that of collagen (94% of mass < 0.0424mm). The solubility of bone broth was found to be much higher than that of egg white. This was found to be the same for both spray-dried and freeze-dried samples. This is expected due to some of the factors affecting the solubility of compounds. It was found that egg white has a higher fat content than bone broth (Table 1). This high-fat content makes the egg white hydrophobic because fatty substances tend to have a lower polarity. Whereas with bone broth, there is a very low-fat content, which is expected to be hydrophilic and polar, making it easily soluble in water.

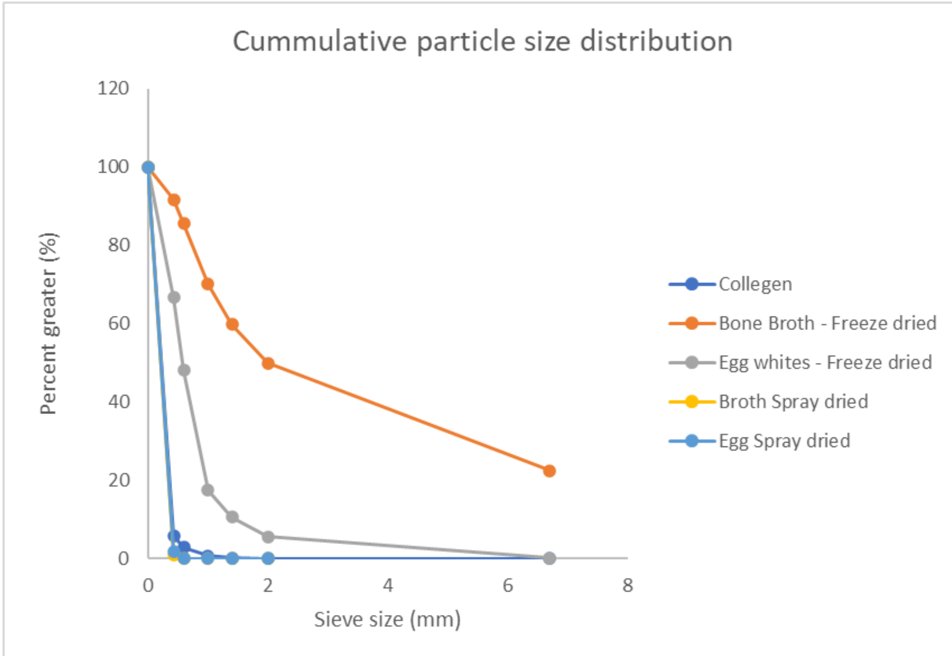

Figure 2: Cumulative Particle size distribution

6. Conclusions recommendations

Food loss is defined as food lost in production or in the supply chain before it reaches the retail level. Waste from the livestock and poultry industry includes unused body parts (bones, horns etc), broken eggs and feathers. Food loss is typically associated with loss of quality or low-quality by-products during the production, processing, and distribution stages of the supply chain. Fourteen per cent of the world's food is lost before it reaches the retail level (Ominski et al., 2021).

6.1. Livestock waste

Slaughterhouses generate solid waste of 27.5%, 17% and 4% of the animal weight for bovine, sheep/goat, and pigs respectively (Mozhiarasi & Natarajan, 2022). The blood from slaughterhouses is one of the major animal byproducts and contains approximately 18% of proteins (Mozhiarasi & Natarajan, 2022). The dry protein could be used to produce yoghurt, cakes, and cheese due to its excellent gelling and emulsifying properties.

6.2. Poultry waste

32.5–37.0% of poultry waste is being generated during the slaughtering of chickens, with the waste composition consisting of 57.37% of feathers and skin; 20.35% of intestines; 14.8% of legs and others (< 1%) (Mozhiarasi & Natarajan, 2022).

Table 8: Waste characterization source Mozhiarasi & Natarajan (2022)

Substrate	**Moisture %**	**TS %**	**VS %**	**Protein %**	**Lipids%**
Poultry trimmings and bones	77.6	22.4	68	11.4	4.9
Cattle meat and fatty waste fractions	47.3	52.7	98.9	6.5	43.2
Bovine slaughterhouse waste	46.8	53.2	98.8	3.5	46.1
Poultry feathers	8.8-12.3	87.7-91.2	85.5-93.5	80	3

6.3. Barriers

Despite the abundance of agricultural waste available, there are several challenges regarding their valorisation and/or preservation.

Several technologies exist for the management of livestock and poultry wastes or for effective product recovery; nevertheless, in most small/medium farms, these technologies are not utilized. Some obstacles prevail, such as space limitations, improper waste segregation at the source, and knowledge of available technologies. Apart from proper waste collection and segregation, financial constraints discourage farm management from perusing waste treatment technologies. Speedy and accurate analysis to determine the nutritional value/quality of by-products and food waste is essential if these dried products from livestock and poultry products are to be utilized consistently in other products.

Economic viability and logistics associated with the collection, transport, and handling of waste may be a significant barrier. especially considering small/medium farmers who may not produce enough waste to warrant adopting processes to valorise or preserve wastes.

6.4. Freeze drying vs Spray drying

From the results obtained in the shelf-life studies, it can be noted that the samples that were spray-dried had the presence and growth of TVC, whereas the Freeze-Dried Bone Broth and Egg White samples showed outstanding results for the TVC, yeast & mould counts. This implies that the spray-dried samples are prone to spoilage faster than freeze-dried samples. Furthermore, the process of freeze-drying was efficient in killing all microorganisms present in the batches of samples that were freeze-dried for this shelf-life study. Our results showed that both spray and freeze-drying had a good distribution of amino acids. However, spray drying leads to the denaturing of complex proteins. The wide range of PSD distribution from freeze-dried products meant that further milling or grinding would be required, which will increase the overall energy requirement of drying broth and egg whites by 1.7 kWh/kg and 0.4 kWh/kg, respectively. The subsequent use of dried products would have to consider the use of additives to improve the solubility, particularly of freeze-dried egg whites, which showed poor solubility potential.

6.5. Recommendations for follow-up action

6.5.1. Explore extraction of protein

The livestock slaughterhouse and poultry byproducts are potential resources for the generation of value-added products, particularly proteins and amino acids. Keratin, a fibrous protein usually found in inedible wastes (hair, nails, feathers, wool, and horns of mammals, birds, and reptiles) could be extracted from wastes for use in pharmaceutical, biomedical, food, and cosmetic industries. Studies have achieved up to 88% of keratin recovery from waste chicken feathers (90% keratin) using sodium sulphide as a reducing agent (Mozhiarasi & Natarajan, 2022). Inedible wastes also contain a high amount of protein that could be extracted for various applications. A study by Selmane et al. achieved a protein yield of 75%, 64%, and 83% for pork lungs, beef lungs, and chicken scraps respectively. These proteins could be valuable additions to meat products.

6.5.2. Test-dried product safety

Further testing for the presence of coliforms and pathogenic Microorganisms (E. coli, Salmonella, Shigella, pseudomonas, Staphylococcus, Bacillus, streptococcus, clostridium, and anthrax), is recommended to confirm the safety of these products samples for human and animal consumptions.

7. Project Limitation

Difficulties in getting financial clearance through the procurement office to timeously analyse our samples, which led to spoilage, delay in achieving tasks on the timeline and inability to extend the scope of the project.

8. Budget

Table 9: Budget used in the project.

Budget heading	Total budget allocated	Total expenditure to date
Site visits	R15,000	R1000
Lab equipment and consumables: glassware, tubing, pipette tips, nitrile gloves etc., including chemicals	R15,000	
Laboratory tests and analysis	R70,000	R56,803.42
TOTAL	R100,000	R57,803.42