Towards more sustainable research: reducing the environmental impact when working with *Drosophila*

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Abstract

The ever-increasing amounts of plastic waste and greenhouse gas emissions worldwide threaten our environment. Biomedical laboratories across the world generate serious amounts of plastic waste often disposed of via high-emission strategies. Achieving sustainability is imperative but requires awareness and knowledge of the regulations, available options and their implications. To illustrate the thought processes involved we showcase the Manchester Fly Facility which supports work with the genetic model organism *Drosophila* and serves 13 research groups. In 2022, we estimated ~4 tonnes of 'clinical' waste generation by the facility enriched with single-use polystyrene plastic containers, all frozen for 2 days and then incinerated. We calculate the resulting environmental and economic costs and compare them to practices reported to us from other fly facilities worldwide. We then discuss feasible management options, separately explaining alternative choices for (1) container materials, (2) the processing of genetically modified organisms, (3) re-use strategies and (4) waste management procedures. This information hopefully raises awareness and understanding to incentivise laboratories worldwide to adopt more sustainable choices, as is permitted by their local infrastructure and regulations. To illustrate what can be achieved, we extrapolate the Manchester data from 2022 to a period of 10 years and calculate the impact of different management strategies, indicating that up to 80% in greenhouse gas emissions and 76% in plastic waste can be saved. The resulting economic savings are of further benefit and could be re-invested to pay for additional workforce, which may otherwise pose an important barrier to re-use scenarios in many countries.

Introduction

Once called “the material of a thousand uses”, plastic has been around for over 100 years owed to its durability, heat resistance and capacity to be moulded into almost anything imaginable (Crespy et al., 2008). However, nowadays plastic use has become a pressing environmental issue: according to the Organisation for Economic Co-operation and Development (OECD), around 400 million tonnes (t) of plastic waste are being generated a year, with its consumption having quadrupled over the past 30 years, and only 9% of this gigantic volume is being successfully recycled (OECD, 2022). Unlike glass, the current plastic recycling options (chemical and mechanical) are limited and rarely promote a circular economy. Moreover, certain recycling methods (e.g. advanced or chemical recycling) are being viewed as environmentally damaging (Hogue, 2022; Open Access News, 2022a).

Of the ~360 million tonnes of plastic disposed of yearly and not recycled, around a quarter is burned and the remainder is sent to landfill (7.2 million tonnes in 2018 in the European Union alone; Wojnowska-Baryła et al., 2022). Unaccounted amounts escape the disposal systems and directly litter the environment. Furthermore, many developed countries export part of their plastic waste to lower income countries, where it is often improperly managed and ends up polluting the air, soil and waters of the entire region (Pandey et al., 2023). At first sight, landfill appears a cheap and convenient method, but some plastics can take around 400 years or more to fully degrade (Chamas et al., 2020). Some of this waste seeps through to the soil and groundwater in the form of microplastics, defined as small pieces of plastic that are less than five millimetres in size (Mortula et al., 2021). By now, microplastics have polluted every part of our planet from the deep oceans (with an estimated 24 trillion pieces of microplastics in our oceans) to the summit of Mount Everest, and it is found in the bodies and blood of animals and humans with expected negative long-term effects on our health (Isobe et al., 2021).

To address this vast ecological and health problem, the UK government has set the goal of eliminating avoidable plastic waste by the end of 2042 through various measures empowered by the Environment Act (UK Government, 2021). The strategy to achieve this goal includes funding of plastic innovation research, the establishment of more efficient recycling systems, the introduction of a tax on plastic packaging that does not contain at least 30% recycled plastic, a ban on key single-use plastic items, such as disposable cutlery or straws, and stricter controls of the exportation of plastic waste (Department Environment Food & Rural Affairs, 2022; Open Access News, 2022b).

Research laboratories must also play their part. Scientists from the University of Exeter estimated in 2014 that biomedical and agricultural laboratories at about 20,500 universities worldwide generated 5.5 million tonnes of plastic waste a year (Urbina et al., 2015). This amounts to ~1.4% of the 400 million tonnes of global plastic waste reported by the OECD in 2022. From our own experience, and talking to colleagues in Biosciences across the world, re-use procedures including cleaning and sterilisation of glass containers, used to be in place in many laboratories, but were later abandoned mostly due to labour costs and unattractiveness of the work involved. Instead, single-use plastic consumables have become ubiquitous over the last three decades, due to their obvious time-saving and cost-effective advantages whilst providing sterility, durability, reliability and low weight (Campion et al., 2025; Farley and Nicolet, 2023; Weber et al., 2025).

The need for action may also be obscured by the wrong assumption that waste incineration is a solution to the problem. However, although incineration can often generate a proportion of energy

from the process, it still is an energy-intense process (Rizan et al., 2021) which also produces poisonous by-products: (1) bottom ash (10-20% of the original weight) is frequently sent to landfill sites where it has the potential to disperse into the environment; it contains heavy metal-coated microplastics and dioxins (Shen et al., 2021; Yang et al., 2021; Zhao et al., 2010; Zikhathile et al., 2022); (2) fly ash (3-5% of the original mass) can be inhaled or contaminate soils through precipitation; it also contains dioxins and heavy metal-coated microplastics as well as high levels of chlorines (He et al., 2023; Shen et al., 2021); (3) gas emissions include nitrogen oxides, sulphur dioxide and carbon dioxide (Aakko-Saksa, 2023) which are a health hazard and display a significant carbon footprint.

To change current practices in research laboratories, the UK "*Concordat for the Environmental Sustainability of Research and Innovation Practice*" published in April 2024 is intended to act as a catalyst (Wellcome Trust, 2024). Furthermore, recent studies have shown that re-use scenarios in laboratories can reduce plastic waste and carbon footprints by impressive amounts (Farley and Nicolet, 2023; Trusler et al., 2024), which should provide a strong incentive. Since research institutions can be expected to be dedicated places of innovation receptive to evidence-based arguments, they might be a good place to address these challenges with innovative solutions, thus serving as trailblazers for other areas of society

However, the considerations and thought processes that might lead to such developments are complex. To illustrate this, we focus here on science laboratories that use, as their research model, the fruit or vinegar fly *Drosophila melanogaster* (and occasionally also related species). *Drosophila* research has a century-long tradition and significantly promoted our understanding of many aspects of biology, with enormous impact particularly on the biomedical sciences (Brookes, 2001/2002; Mohr, 2018; Prokop, 2018). *Drosophila* research is carried out in ~2000 laboratories across the globe (FlyBase Wiki!, 2025) where fly stocks are usually kept in large numbers in glass or plastic vials/bottles. Many fly stocks classify as genetically modified organisms (GMOs), the husbandry and deposition of which is regulated. The containers are being re-used in some places, but most places appear to practice single-use, thus generating large amounts of plastic waste accompanied by a high carbon footprint as plastic is made from fossil fuel and its production requires energy.

Here we take the Manchester Fly Facility as one example of waste management relating to genetic or biomedical research. We characterise the waste produced, apply established analysis methods to measure the environmental impact of current waste management strategies, compare them to procedures at other fly facilities and then explain the pros and cons of available single-use and re-use strategies and how they can be combined in a modular fashion. We hope this will help and incentivise research laboratories worldwide to implement sustainable practice according to the local requirements and possibilities.

Methods

Our case study is based on a project carried out in 2022 at the Manchester Fly Facility which established the number of containers and estimations of equipment usage times during the entire year. Greenhouse gas emission calculations were based on the newest and, in our view, best-established data suggested in publications (details below). Costings of material, energy and services

were adapted to the latest available prices when writing this manuscript (see Tabs. 1 and 2). The calculations are provided in a supplementary file.

Greenhouse gas emission calculations

Greenhouse gas emissions are measured in carbon equivalent (CO_2eq). CO_2eq is a metric measure comparing the emissions from various greenhouse gases by converting their amounts into the equivalent amount of CO_2 with the same global warming potential (eurostat, 2023)

- Glass containers: we used the standardised GHG conversion tables for company reporting published annually as spreadsheets by the UK Government, here forth referred to as government coefficient (Departments for Energy Security and Net Zero and for Business Energy & Industrial Strategy, 2023). The coefficient for glass (1.4 g CO_2eq/g) includes the extraction, primary processing, manufacturing, transportation of each material to its point of sale. As glass is heavy, it is important to include transportation.
- Plastic containers: for polystyrene (PS) and polypropylene (PP), we compared the production coefficients provided by the government to those recently published in a meta-analysis for single-use plastic lab consumables (Ragazzi et al., 2023). For PS, the values were the same (3.8 g CO_2eq/g). For PP, the government coefficient was 3.09 g CO_2eq/g, whereas Ragazzi and co-authors used 2.5 g CO_2eq/g (based on 4 values within the range of 2.1 and 2.7); they also argued that transport and packaging produced negligible emissions and can be ignored. We decided to use the 2.5 g value as it seemed better substantiated.
- Electricity: for the running energy cost of freezers, washer/dryers and autoclaves (for calculations of usage see below), we used the coefficient for the UK grid provided by the government (0.2 kg CO_2eq/kWh; Departments for Energy Security and Net Zero and for Business Energy & Industrial Strategy, 2023). Further emissions arise from water supply and treatment estimating ~1% for autoclaves and ~5% for washers (Farley and Nicolet, 2023)
- Landfill waste: we referred to the government coefficient for sending materials to landfill. our waste composition (plastic, fly food, blue roll, cotton/celluloid plugs) best matched their house residual waste category (500 kg CO_2eq/t); this value is high because the food, paper and cotton plugs produce CO_2 during aerobic decomposition and a mixture of CO_2 and methane in anaerobic conditions over a long period of time (Department for Environment Food & Rural Affairs, 2022). Of these, methane has a global warming potential which is 28 times higher than CO_2 over 100 years (Box 3.2 on p.103 in Intergovernmental Paen on Climate Change, 2014); this explains the high landfill emission coefficient.
- Incineration and autoclaving: the government coefficient factors only account for transport and are therefore insufficient. Since we do not have access to the emissions data from our local waste facilities (Oldham Clinical Waste Incinerator run by Stericycle Ltd), we based our estimation on recently published coefficients for various waste streams at a UK hospital site, which seemed comparable to our institution (Rizan et al., 2021). For the biosafety treatment by autoclave, we used their breakdown of 75 kWh of electricity, 92.17 litres of gas oil and 1.93 m3 of water ((270 kg CO2eq/t); we adapted the travel distance for the low temperature incineration (850°C) with energy recovery (175 kg CO_2eq/t); for the high temperature incineration of clinical waste (1000 °C), our local facility does not quantify the degree of energy recovery (not granted an R1 code), so we

estimated the energy recovery based on that for low temperature incinerator reported elsewhere (170 kg CO_2eq/t; Rizan et al., 2021) and adjusted the travel distance, resulting in a coefficient of 785 kg CO_2eq/t. This said, the LCA database by Ecoinvent uses the much higher value of 2570 kg CO_2eq/t for clinical incineration according to (Ragazzi et al., 2023), so our values may only represent a third of the actual values.

Costings

- For the containers, we referred to suppliers' websites in June 2024.
- For electricity, gas and oil, water supply and treatment, we referred to the rates reported by AquaSwitch (AquaSwitch) for June 2024 for large businesses: electricity (20.78 p/kWh), gas and oil (5.2 p/kWh), water supply (2.07 £/m^3) and water treatment (1.51 £/m^3).
- For waste, our waste coordinator provided an estimated cost for incineration of £270/t for the low temperature incineration and £630/t for the high temperature incineration in May 2023. We have a no-landfill policy at our Institution, but the cost of sending waste to landfill would be expected to be ~£160/t.
- Salary costings for container re-use: To empty the containers and put them in baskets for the washer, we estimate manual labour of 30 mins for 100 vials based on empirical data at Oxford. The processing of 10,167 containers a month would thus equate to 51 hrs/month and 6,100 hrs over 10 years. Working hours in the UK are typically 140 hrs/month, so we would need technical support staff paid at 0.3 FTE grade 3. If we assume entry level, the annual salary is £24,248 and the hourly rate is £13.32. The manual labour would thus cost £81,255 over 10 years.

Cycle run and cost calculations for the use of freezers, washers, driers and autoclaves

The kWh usages of existing local appliances were obtained by contacting the manufacturers, and the calculated CO_2eq values are listed in Tab. 3. Our two Esta freezers (no longer produced) can hold 3 waste bags containing several hundred vials or ~50 bottles each; each freezer consumes 2 kWh per day and are left on continuously (0.8 kg CO_2e and £0.83 per day), so that the waste turnover is the same for single- and re-use. Our Lancer 1300LX washer and dryer can hold six 125 mm x 425 mm trays providing space for 438 vials and 8 bottles per cycle. Given a requirement for 10,000 vials and 167 bottles per month, we would need a minimum of 23 cycles/month. The programme considered washes for 1 hour at 95°C and dries at 85°C, consuming 4.5 kWh per cycle (0.9 kg CO_2e and £0.93 per cycle). The existing autoclave (Vakulab PL H-Model 969) has a capacity for 2,500 vials and 48 bottles per cycle, so we need a minimum of 4 cycles a month; it maintains the temperature at 121°C for 30 min and consumes 22.5 kWh per use (4.5 kg CO_2e and £4.68 per use). To allow for potential extra demand, our calculations considered 25 cycles/month for the washer/dryer and 5 cycles/month for the autoclave if used after each wash. For the sporadic scenario (e.g. autoclaving only upon stock contamination) or precautionary intercalated autoclave cycles, we anticipate a maximum of 2 cycles/month.

Calculating container numbers for re-use scenarios

According to experiences at Oxford, PP containers can be re-used at least 20 times (see case study) and we should account for a 10% loss over the years. If vials can be re-used 20 times at a loss of 10%

per year, this means that we need 6.6 months (132/20) of supply upfront to last 10 years, i.e. 66 000 vials and 1100 bottles.

Determining the amount of waste for single-use and re-use scenarios.

The amounts of bags disposed of each week within the current single-use scheme was monitored and their weight taken for a period of 4 weeks. The product of average weight of bags multiplied by their average number over this period was calculated and extrapolated to 52 weeks to establish the yearly average (note that fly stock maintenance requires uninterrupted cycles of vial turn-over so that no holiday periods were considered in our calculations). This amounted to 4 t of waste in 2022 for the single-use scenario. For the re-use scenario, we subtracted the weight of the plastic containers consumed in 2022 (number of vials and bottles used x their weight = 0.76 t) from the total waste, giving us a total of 3.2 t of annual waste, 2.2 t of which are disposed fly food (number of vials and bottles used x average amount of fly food per container: 17 g for vials and 105 g for bottles).

Results

Current practices at the Manchester Fly Facility

The Manchester Fly Facility is situated at the Faculty of Biology, Medicine and Health of The University of Manchester and serves 13 research groups using the genetic model organism *Drosophila melanogaster* for their research (Manchester Fly Facility, 2014). It generates considerable amounts of waste. Incentivised by the university's move to implement the Laboratory Efficiency Assessment Framework (LEAF) to promote sustainable practice and its intramural 6R strategy to reduce plastics (Farley, 2022; Ow et al., 2025; Taylor-Hearn, 2023; Wattam et al., 2024), we investigated the environmental costs of our current practices and explored alternative strategies.

As explained in greater detail elsewhere (Ashburner et al., 2005; Roote and Prokop, 2013; Stocker and Gallant, 2008), work with *Drosophila* usually requires the husbandry of hundreds of fly stocks per research group, each displaying a distinct genetic modification. Each *Drosophila* stock is usually maintained in copies of two vials stored in trays that are placed in 18°C incubators or temperature-controlled rooms (Fig.1A). Flies from each vial need to be tipped onto fresh vials every four weeks. For experimental purposes, flies are boosted in larger amounts using vials or bottles that are kept at 25°C. At this temperature, the developmental time is roughly half that of 18°C storage requiring container turn-over every fortnight.

At the Manchester Fly Facility, *Drosophila* stocks are kept in polystyrene vials (25 mm x 95 mm) or polypropylene bottles (43 mm x 132 mm; see Tab.1 for details) which are plugged with cotton wool or celluloid plugs and carry, at the bottom, a several centimetre-high layer of food composed of yeast, glucose, agar, maize, propionic acid and nipagin (for recipes see Bloomington *Drosophila* Stock Center, 2021; Fig.1C). Bottles are delivered by the local media kitchen in plastic sacks and vials in cardboard trays holding 100 units sealed into a plastic bag (Fig.1B).

For the year 2022, we performed a detailed analysis establishing that the Manchester Fly Facility generated 120,000 polystyrene (PS) vials and 2,000 polypropylene (PP) bottles in waste. These containers are discarded into plastic bags (weighing on average 9 kg) and placed in a freezer at -20°C

for 48 hrs to ensure that flies and mites at all developmental stages have been killed, as is the locally agreed procedure for genetically modified organisms. Thereafter, the bags with their frozen contents are put into large yellow clinical waste bags (Fig.1E) which are sent to the Oldham clinical waste incinerator (Fig.1F; see Methods), following guidance for biohazardous waste by the Scientific Advisory Committee on Genetic Modification (SACGM; Health & Safety Executive). The total annual waste created by the Fly Facility in 2022 (see Methods) was ~4 tonnes, of which ~760 kg (~20%) are plastic vials and bottles and the rest comprises ~2.2 tonnes of fly food and fly carcasses and ~ 990 kg of miscellaneous items such container plugs and paper towels (Fig.1). The non-contaminated plastic bags used to package fresh bottles or vial trays can either be re-used or recycled via our soft plastic recycling stream (https://www.elsarecycle.co.uk). In the following sections, we will calculate the carbon footprint and costs of the current procedures and compare them to alternative strategies.

Cost analysis of existing and feasible other single-use scenarios at the Manchester Fly Facility

Single-use practices involve making a choice of material for the vials, of the method to comply with genetically modified organism regulations, and of the disposal method. To establish the carbon footprints of these various components, we calculate their greenhouse gas emissions measured in carbon equivalent (CO_2eq), as is explained in the first Methods section. For our CO_2eq calculations (see Methods), we used our measured consumption data for the year 2022 (Fig. 1: 120,000 vials, 2,000 bottles, 4 t total waste) and extrapolated them unchanged to a period of 10 years. This long-term view makes it easier to compare single-use with the re-use scenarios that will be discussed further down in this article. We calculated the costs involved using the newest data available up to June 2024 for a more accurate picture at the time of publication, as energy and consumables prices have been very volatile in recent years (see Methods).

For plastic containers, we calculated their *cradle to gate* costs, i.e. the CO_2eq emissions associated with material production and manufacturing of the containers, whereas transport and packaging was negligible. These calculations showed that PP is a better choice than PS, saving 9.3 t CO_2eq over a decade. Currently, PP is more expensive than PS (an extra £24,000 over 10 year), but prices are fluctuating and depend on volumes traded. Glass has not been considered in the single-use scenario due to its high purchase cost (Tab.1).

For GMO compliance procedures, we considered autoclaving or 48 hrs freezing at -20°C. Of these, freezing flies is far more sustainable than autoclaving, saving 7.9 tCO_2eq over 10 years at a similar economical cost (Fig.2).

For disposal, we considered different options. One disposal option in the single-use scenario is waste incineration. It is common standard in the UK to incinerate decontaminated or non-hazardous offensive waste at low temperature (~850°C) and clinical or hazardous waste at high temperatures above 1000°C (highT; Rizan et al., 2021). Both procedures are energy intense even if the incineration process is used to recover energy, referred to as energy from waste (EfW; as is the case at Manchester: see Methods). HighT incineration with EfW of the 40 t of waste over a decade amounts to 31.4 t CO_2eq and an economical cost of £25,200. Compared to this, lowT incineration with EfW would save 24.4 t CO_2eq and £14,400 over 10 years. The other option is landfill which would save £18,800 in economical cost but only 11.4 t in CO_2eq cost due to the production of methane which is a very potent

greenhouse gas (Department for Environment Food & Rural Affairs, 2022; Intergovernmental Paen on Climate Change, 2014; see Methods). Furthermore, landfill damages the environment through the generation of harmful microplastics (Wojnowska-Baryła et al., 2022). Therefore, our data indicate lowT/EfW to be the most sustainable and hightT/EfW the most wasteful of these three scenarios.

When combining GMO treatment and disposal, the best options appear to be freezing followed by lowT incineration (9.9 t CO_2eq) or autoclaving followed by lowT incineration (~17.8 t CO_2eq). Therefore, even if GMO officers insisted on autoclaving, the use of lowT would still save 16.5 t CO_2eq over freezing combined with highT and 13 t CO_2eq over autoclaving plus landfill when calculated over a decade.

Practices at other fly facilities

To establish how other institutions handle fly husbandry and learn about viable alternatives, we consulted four UK-based fly facilities from Cambridge, Oxford, Bournemouth and London, one from India (Bangalore) and the Bloomington *Drosophila* Stock Center (Indiana, US). We gathered information via a survey asking about materials used for vials and bottles, whether single-use or re-use of these materials was implemented, and the employed disposal methods (summarised in Tab.4).

The survey confirmed that the three above-mentioned materials (PS, PP, glass) are being used for fly containers worldwide. Some fly facilities had a single-use system in place, others re-use. To comply with GMO regulations, freezing and/or autoclaving are being used. Waste disposal strategies include landfill, lowT and highT incineration, as well as release of food and dead flies into the sewer (details in Tab.4).

To illustrate the steps involved in re-use procedures, we provide two protocols. The first example is currently in practice at the Raff laboratory in Oxford, UK (Box 1, Fig. 3). It involves manual cleaning of PP containers, incineration of plugs and organic waste, and chemical disinfection with a 1% solution of Chemgene HLD4L. This laboratory disinfectant is effective against bacteria, fungi, mycobacteria and viruses (with few limitations; Kampmann et al., 2024; Uy et al., 2022), and safety data sheets by vendors indicate little ecotoxicity (although we could not find any published evidence). However, the experience in Oxford shows that researchers still prefer autoclaving as the best-proven method of decontamination to prevent spread of potential infections in fly stocks. Therefore, longer-term tests would be required to build trust in disinfection with Chemgene.

If autoclaving stays the option of choice in re-use scenarios, it should be considered to leave out the chemical disinfectant during the cleaning step (Box 1) potentially replacing it with ecologically friendly detergents. Also, PS vials cannot be used because they are less heat-resistant than PP (heat deflection temperature is 107°C for PP and 82-96°C for PS; ThermoFisher). Experience in Oxford has shown that PP containers can be autoclaved and re-used at least 20 times (Fig.3L). This said, PP vials are slightly less transparent than PS (Fig.3L) and glass vials might be an alternative, although glass can splinter upon impact causing issues including cuts or escape of flies potentially affecting ongoing experiments.

Box 1: Example of a re-use protocol as established at Oxford

1. **Collection:** Collect used *Drosophila* plastic vials and bottles in a container until it is full (Fig.3A).
2. **Freezing:** Freeze the collected vials and bottles at -20°C (or lower) for at least 48 hrs to ensure that flies and mites at all stages of development are killed.
3. **Thawing:** Thaw vials and bottles at room temperature (RT) for at least 2 hrs.
4. **Removing Plugs:** Separate the plugs from the vials and bottles and discard the plugs in a waste box (Fig.3E; see item 6)[A].
5. **Soaking:** Soak the vials and bottles in tap water for at least two hours to soften the organic contents and facilitate their removal (Fig.3B).
6. **Removing Food Waste:** Use a spatula to remove organic contents and collect in a sieve, letting surplus water escape down the sink (Fig.3C,D); organic material captured in the sieve can be autoclaved and disposed together with the discarded plugs[B].
7. **Cleaning:** Emptied containers (Fig.3F) are submerged in a 1% solution of the disinfectant Chemgene HLD4L Conc ; in this solution, use a bottle brush with a cloth tip to scrub the inside and outside of the containers (Fig.3G).
8. **Soaking in Chemgene:** After cleaning, keep the vials and bottles in Chemgene solution for another 30 minutes to achieve proper decontamination (Fig.3H).
9. **Rinsing:** Rinse thoroughly with tap water, followed by a final rinse with distilled water (Fig.3I) and rub off labels with alcohol (Fig.3J).
10. **Autoclaving (optional):** Autoclave the vials and bottles (to deal with potential resistances against Chemgene)
11. **Storage/Refill:** Place the containers on trays ready for refill with fly food (Fig.3M).

i. **Re-use:** Unless mishandled, vials typically withstand more than 20 cycles of wash and re-use (more for bottles; Fig.3L); continue re-using the vials and bottles until signs of wear are evident, such as cracks or reduced transparency.

ii. **Time Requirement:** The total time taken to wash and regenerate 100 vials or bottles is approximately 1 hr.

A: Some groups use plastic stoppers which can be washed, autoclaved and re-used.

B: You may be able to skip autoclaving if the litter is sent to incineration (see main text).

The second example of a re-use scenario describes procedures at the Bloomington *Drosophila* Stock Center with an annual volume of ~4.4 million vials, hence designed for high efficiency whilst following NIH guidelines (see Discussion). This scenario involves 10 min autoclaving, a 30 min dishwasher step and 1hr drying at 80°C (Box 2).

Box 2: Re-use procedure used at the Bloomington *Drosophila* Stock Center (~10 min labour time)

1. For stock maintenance, mostly glass vials with rayon plugs (*made from natural cellulose*) are used; dirty plugged vials are placed upright in galvanised metal trays of ~250 vials using racks that hold the trays at an angle to facilitate the task.
2. Trays are autoclaved, not to sterilise the vials but to kill the flies and melt the agar using an abbreviated autoclave cycle (*a gravity cycle of ~8 minutes at full heat and pressure*).
3. Plugs are removed and sent to landfill.
4. Molten vial contents are rinsed out with a hose into the sewer.
5. Vials are cleaned for ~30 minutes in a lab dishwasher with a bleach-containing, high alkaline cleaner (*similar to those used in dairies and ideal to sterilise the vials; no further autoclaving is used*).
6. Vials are transferred to a glass-drying closet at ~80°C until dry (*~1 hr*). They are then ready for re-use.

Broken glass or old plastic vials are frozen over night (*from then on no longer considered biohazardous waste*) and sent to landfill.

Cost analysis of potential future re-use scenarios at the Manchester Fly Facility

In the following, we will assess potential re-use scenarios for the Manchester Fly Facility, based on procedures reported by other fly facilities (Tab.4, Box 1), the infrastructure available at the Manchester Fly Facility, and the projected use of 1.2 million vials and 20,000 bottles in a decade.

First, we calculated how much stock must be bought in to sustain the re-use scenario. In our calculations, we considered that PP containers have a lifetime of ~20 cycles and assume ~10% loss of containers every year due to ageing and damage (Farley and Nicolet, 2023; above-mentioned experiences at the Raff laboratory). Considering these various parameters, we calculate that a total of 66,000 vials and 1,100 bottles needs to be purchased during the 10-year period (see Methods). The total waste including food, fly carcases and discontinued vials is expected to amount to 3.2 t (see Methods).

Like in the single-use scenario, the first step of the procedure is freezing at -20°C for 48h to kill flies. The second step would be the manual removal of stoppers and organic contents from the vials which need to be disposed off (potentially requiring an autoclaving step first) either using conventional landfill or incineration (lowT or highT); disposal to biogas facilities or into the sewer is currently not an option at Manchester. Potential plastic stoppers could be washed and sterilised for re-use (not considered here). In the next step, empty containers need to be cleaned, either manually or using machine-washing at 95°C, and decontaminated using chemical disinfection or autoclaving; containers may be autoclaved in each re-use cycle, or only occasionally when there are contamination issues (here conservatively calculated at twice a month instead of 5 times a month). Fig. 2B summarises the calculated costs and CO_2eq emissions for the individual components of the re-use scenario (obligatory and optional) over a period of 10 years (see Methods for details of calculations).

Unsurprisingly, the footprint associated with material production for PP vials is reduced by almost 20-fold when assuming 20 re-use cycles (Fig.2B). Using glass has only a slightly higher footprint than PP when considering re-use scenarios (Fig.2B), in contrast to single-use scenarios where the cost would

be astronomical (£509,670 and 37.1 t CO_2e; Tab.1). Furthermore, the data illustrate that washing steps raise the costs only marginally, even when using machine wash (Fig.2B); it is rather the labour cost for emptying containers and then handwashing or loading machines which needs careful consideration in cost/benefit calculations (see Discussion). Key differences arise from decisions about the necessity for autoclaving the waste (Fig.2B), for example whether local GMO officers can be convinced that autoclaving is an unnecessary step if flies have been killed by freezing and waste is incinerated anyway. With respect to incineration, the choice of lowT over highT is of enormous further benefit.

As should have become clear from all the above considerations and data, waste management strategies can be assembled in flexible ways. Key choices should consider the material of containers (PP being preferred), GMO procedures (freezing being preferred), re-use versus single-use (re-use preferred, but workload needs careful consideration), and methods of waste disposal (lowT incineration of plastics and disposal to biogas or composting facilities preferred). Researchers should consider their local infrastructure, conditions and regulations and strive to choose the combination within these limitations that has the highest ecological benefit.

Discussion

A modular approach to sustainability providing opportunities and unexpected insights

The intention of this study was to assess the feasibility and beneficial impacts of more sustainable waste and resource management strategies at the Manchester Fly Facility, with a view to inspiring *Drosophila* research groups worldwide to review their practices, and hopefully also other laboratories that use large amounts of plastics and generate biological waste. The key idea of our approach was to consider single-use and re-use scenarios and to break them down into their individual compulsory and optional or alternative components (Fig.4). By discussing the cost and benefit of each of these choices, we aimed to provide a list of modules covering different material options, potential cleaning avenues, as well as alternative GMO and disposal procedures. The information provided for these modules should allow research groups to work out combinations that will lower the environmental burden and are feasible in the context of the respective local infrastructure. Examples of different combinations will be discussed in the next section.

Work on this article provided interesting new insights. Firstly, we had a preconception that glass containers were the only feasible option for re-use scenarios because they remain fully transparent even after many cleaning cycles. However, we learned that also PP vials withstand at least 20 re-use cycles in good condition providing a viable alternative at comparable CO_2eq cost to glass, whilst being cheaper to purchase and less prone to break if accidentally dropped during experimental or stock maintenance work or when cleaning for re-use (Fig.2B).

Secondly, we learned about the enormous difference in CO_2eq emissions between highT versus lowT incineration. We already used this argument here at Manchester to get our waste re-classified from ‘clinical’ waste requiring highT, to ‘offensive’ waste that can be sent to lowT incineration (National Health Service, 2023).

Thirdly, we now strongly argue against autoclaving steps before waste disposal. As long as flies have been killed, autoclaving adds substantial CO_2eq cost and workload without any additional benefit for

biosafety. At the Centre for Neural Circuits and Behaviour in Oxford, freezing of flies is followed by lowT incineration as standard practice. In the US, standard transgenic and mutant *Drosophila* fly stocks are handled under the NIH guidelines for biosafety level 1 (BL1) and must be rendered 'biological inactive before disposal'; for arthropods this means they can be 'killed with hot water or freezing before flushing down drains or placed into trash bags' (American Committee of Medical et al., 2019; NIH Guidelines, 2024).

Fourthly, we considered microplastic contamination as the major issue of landfill but were surprised to learn about the enormous CO_2eq burden in conventional landfill scenarios where anaerobic decomposition of organic and paper waste generates high amounts of methane (Fig.2). This clearly argues against the use of conventional landfill.

Through work on this article additional ideas arose. For example, the collected organic waste at Manchester amounts to 2.2 t a year (50% of our total waste; see Methods for calculations) which is currently incinerated at highT. An alternative strategy would be to use it for compost production or in biogas plants which are carbon-neutral or even -reducing options (Nordahl et al., 2020). Potential interference of anti-fungal or anti-bacterial compounds in the fly food with natural processes of decomposition could be avoided by heat treatment (propionic acid has a flash point of 60°C). The University of Manchester already sends food waste to a biogas plant. If switching to a re-use scenario, we could join this stream, thus further reducing the incinerated waste from 32 t to 10 t over 10 years, lowering the environmental impact by 2/3 and likely also the economic cost. In locations where local wastewater purification plants use the sewage sludge to feed biogas production (Sever and Decorte, 2024), disposal of organic waste into the sewer might achieve the same effect whilst further lowering costs and work. Two fly facilities we contacted already use disposal of food and dead flies into the sewer as standard procedures (Tab.4).

In conclusion, there are several options to be considered, and choices will have to be made based on local infrastructure and GMO rules - of which the latter might be addressed through constructive discussions.

Comparing different combinations of waste management modules

To illustrate the implications of decisions taken when designing waste and resource management strategies, we compare four different scenarios (Fig. 5). The first scenario (‘single 1’ in Fig.5) is currently used at the Manchester Fly Facility with a very high environmental cost: it uses PS vials, PP bottles, freezing, and highT incineration (Fig.1), summing up to 62.6 t CO2eq and £156,014 over 10 years (keeping prices constant).

The second scenario (‘single 2) is the one we aim for as long as we continue with single-use: change from PS to PP vials, and from highT to lowT, ideally avoiding autoclaving. The latter two aspects are being discussed with local GMO officers, and our main argument would be a reduction in environmental cost over 10 years of more than 50% down to 29 t CO2eq compared to scenario 1. When considering the economic cost, scenario 2 is currently ~10% more expensive than scenario 1, mainly due to the higher cost for PP vials compared to PS. This said, production costs may come down upon increased sales volumes if more fly facilities would change to PP vials. Clearly, the

economic cost of purchasing containers is the dominating cost in single-use scenarios, which is up to 15 times higher than in re-use scenarios (Fig.5).

The first re-use scenario ('re-use 1') illustrates the impact of landfill. It involves re-use of vials following machine-wash and autoclaving of vials and autoclaving organic waste before sending it to landfill. Compared to single-use scenario 1, re-use 1 offers a reduction of ~46% in CO2eq cost and ~84% in financial cost (excluding labour cost). The key CO_2eq burden in the first re-use scenario is generated by the autoclaving step and the use of landfill. To improve on these shortcomings, the second re-use scenario ('re-use 2') considers disposal via lowT incineration and assumes that the autoclaving of organic waste can then be omitted (whereas autoclaving of containers may be demanded by users and has been included in Fig.5). Compared to single-use scenario 1, re-use 2 offers a reduction of 76% in environmental cost and 84% in economic cost (excluding labour cost).

All re-use scenarios would involve the manual removal of organic waste from vials following the freezing step. If cleaning 100 vials with a spatula takes up to 30 minutes, this would equate to ~6,100 hrs for 1.22 million containers (vials and bottles). At Manchester, paying a technical assistant at the adequate grade for 10 years would amount to ~ £81,000 (not factoring in salary increases; see Methods) which would be comfortably covered by savings on other components: for example, changing from the current single-use scenario 1 to re-use scenario 2 would save ~£130,000 over 10 years (Fig.5). However, similar calculation at any fly facility will have to take into consideration what infrastructure (e.g. washers and autoclaves) at the necessary capacity is locally available for re-use scenarios or needs to be purchased, whether running costs deviate from those of the appliances used for our calculations, how equipment is maintained, and the local price of paid labour which will differ considerably.

Future perspectives

As stated before, there are an estimated 2000 fly facilities worldwide (FlyBase Wiki!, 2025) which can use the information provided here to consider local practices and engage in discussions with colleagues, local managers and GMO officers: for example, to change from high-CO_2eq disposal (landfill or highT) to more sustainable lowT incineration, or to invest in workforce to implement re-use scenarios, thereby reducing plastic production and microplastics release. But we should not stop here, and strategies and arguments must be further refined and strengthened. For example, we should consider sustainable decomposition procedures of organic material (see above) but also recycling of broken or old plastic containers (the fly lab in Oxford already uses a contractor to this end). New materials should also be considered, including recycled PP or biodegradable plastics, provided they withstand the heat of newly cooked fly food or even autoclaving - which may take considerable time given the state of technology and the competition for the necessary resources (Ragazzi et al., 2023). Further factors not mentioned so far need to be considered, for example reducing water consumption or replacing environmentally damaging chemical substances used in washers. Furthermore, most if not all carbon emission coefficients presented here will change when more green energy is provided by the UK grid, thus improving the environmental costs of all those processes that depend on electricity.

However, the key to success will be that we as researchers and citizens accept our responsibility and necessity to have to go an extra mile for sustainability, despite us being overloaded with work and demands. It is important that those of us who have started to engage on this path then also share their good practices and raise awareness of any new opportunities, for example by talking to colleagues or giving presentations to local audiences or on conferences or by making sustainability an active part of the student learning experience (Smith et al., 2025). Once it becomes a community effort, workload can be shared and synergy achieved that facilitates implementation and will likely also enhance effectiveness. Given that *Drosophila* research has a strong track record of being at the forefront of new developments, we should take on sustainability in the same way and act as trailblazers motivating colleagues at our institutions to follow suit.

Data availability statement

All data are made available through the article and supplementary material.

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Competing interests

The authors have no competing interests to declare

References

American Committee of Medical, E., American Society of Tropical, M., Hygiene (2019) Arthropod Containment Guidelines, Version 3.2. *Vector-Borne and Zoonotic Diseases* **19,** 152-173 -- https://doi.org/10.1089/vbz.2018.2431

AquaSwitch Business electricity prices. *Last accessed:* -- https://www.aquaswitch.co.uk/business-electricity-prices/

Ashburner, M., Golic, K. G., Hawley, R. S. (2005) *Drosophila*: a laboratory handbook. 1409 -- https://archive.org/details/drosophilalabora0000ashb

Bloomington *Drosophila* Stock Center (2021) Fly food recipes. *Last accessed:* 01 June -- https://bdsc.indiana.edu/information/recipes/index.html

Brookes, M. (2001/2002) Fly: The Unsung Hero of Twentieth-Century Science. Ecco/Phoenix, -- https://tinyurl.com/y2ub6l8n

Campion, C., Robertson, L., Stansfield, I., Speirs, V. (2025) Towards greener and more sustainable pre-clinical oncology research. *BJC Reports* **3,** 4 -- https://doi.org/10.1038/s44276-024-00115-0

Chamas, A., Moon, H., Zheng, J., Qiu, Y., Tabassum, T., Jang, J. H., Abu-Omar, M., Scott, S. L., Suh, S. (2020) Degradation rates of plastics in the environment. *ACS Sustain Chem Eng* **8,** 3494-511 -- https://doi.org/10.1021/acssuschemeng.9b06635

Crespy, D., Bozonnet, M., Meier, M. (2008) 100 years of bakelite, the material of a 1000 uses. *Angew Chem Int Ed* **47,** 3322-8 -- https://doi.org/10.1002/anie.200704281

Department Environment Food & Rural Affairs (2022) UK leads the way on ending plastic pollution *Last accessed:* -- https://www.gov.uk/government/news/uk-leads-the-way-on-ending-plastic-pollution#

Department for Environment Food & Rural Affairs (2022) Experimental statistics on the carbon impact of waste from households manages by local authorities in England. -- https://assets.publishing.service.gov.uk/media/63974500e90e077c329444f0/Statistics_on_carbon_emmisions_Waste_Households_England_v8_2018.pdf

Departments for Energy Security and Net Zero and for Business Energy & Industrial Strategy (2023) Research and analysis - Greenhouse gas reporting: conversion factors 2023. *Last accessed:* 16/08/2023 -- https://www.gov.uk/government/publications/greenhouse-gas-reporting-conversion-factors-2023

eurostat (2023) Glossary: carbon dioxide equivalent *Last accessed:* 16/08/2023 -- https://ec.europa.eu/eurostat/statistics-explained/index.php?title=Glossary:Carbon_dioxide_equivalent#:~:text=A%20carbon%20dioxide%20equivalent%20or

Farley, M. (2022) How green is your science? The race to make laboratories sustainable. *Nat Rev Mol Cell Biol* **23,** 517 -- https://doi.org/10.1038/s41580-022-00505-7

Farley, M., Nicolet, B. P. (2023) Re-use of laboratory utensils reduces CO2 equivalent footprint and running costs. *PLoS ONE* **18,** e0283697 -- https://doi.org/10.1371/journal.pone.0283697

FlyBase Wiki! (2025) FlyBase:Fly Lab List. *Blog post* -- https://wiki.flybase.org/wiki/FlyBase:Fly_Lab_List

He, D., Hu, H., Jiao, F., Zuo, W., Liu, C., Xie, H., Dong, L., Wang, X. (2023) Thermal separation of heavy metals from municipal solid waste incineration fly ash: A review. *Chem Eng J* **467,** 143344 -- https://doi.org/10.1016/j.cej.2023.143344

Health & Safety Executive Guidance from the Scientific Advisory Committee on Genetic Modification. *Last accessed:* -- https://www.hse.gov.uk/biosafety/gmo/acgm/acgmcomp/

Hogue, C. (2022) Chemical recycling of plastic gets a boost in 18 US states—but environmentalists question whether it really is recycling. *Chem Eng News* **100** -- https://cen.acs.org/environment/recycling/plastic-recycling-chemical-advanced-fuel-pyrolysis-state-laws/100/i17

Intergovernmental Paen on Climate Change (2014) Climate change 2014: synthesis report - contribution of working groups I, II and III to the 5th assessment report of the IPCC. *IPCC*, -- https://archive.ipcc.ch/ipcc_languages_e_master.shtml#tabs-3

Isobe, A., Azuma, T., Cordova, M. R., Cózar, A., Galgani, F., Hagita, R., Kanhai, L. D., Imai, K., Iwasaki, S., Kako, S. i., Kozlovskii, N., Lusher, A. L., Mason, S. A., Michida, Y., Mituhasi, T., Morii, Y., Mukai, T., Popova, A., Shimizu, K., Tokai, T., Uchida, K., Yagi, M., Zhang, W. (2021) A multilevel dataset of microplastic abundance in the world's upper ocean and the Laurentian Great Lakes. *Microplast Nanoplast* **1,** 16 -- https://doi.org/10.1186/s43591-021-00013-z

Kampmann, M. L., Tfelt-Hansen, J., Borsting, C. (2024) Cleaning protocols in forensic genetic laboratories. *Int J Legal Med* -- https://doi.org/10.1007/s00414-024-03232-0

Manchester Fly Facility (2014) Manchester Fly Facility: supporting *Drosophila* research. *Last accessed:* 01 June -- https://sites.manchester.ac.uk/fly-facility

Mohr, S. E. (2018) First in fly - *Drosophila* research and biological discovery. -- http://www.hup.harvard.edu/catalog.php?isbn=9780674971011

Mortula, M. M., Atabay, S., Fattah, K. P., Madbuly, A. (2021) Leachability of microplastic from different plastic materials. *J Environ Manage* **294,** 112995 -- https://doi.org/10.1016/j.jenvman.2021.112995

National Health Service (2023) NHS clinical waste strategy. pp. 27 -- https://www.england.nhs.uk/long-read/nhs-clinical-waste-strategy/

NIH Guidelines (2024) NIH guidelines for research involving recombinant or synthetic nucleic acid molecules *National Institute of Health*, -- https://osp.od.nih.gov/wp-content/uploads/NIH_Guidelines.pdf?highlight=%22NIH+GUIDELINES+FOR+RESEARCH+INVOLVING+RECOMBINANT+OR+SYNTHETIC+NUCLEIC+ACID+MOLECULES%22&_rt=MnwxfG5paCBndWlkZWxpbmVzIGZvciByZXNlYXJjaCBpbnZvbHZpbmcgcmVjb21iaW5hbnQgb3lgc3ludGhldGljIG51Y2xlaWMgYWNpZCBtb2xlY3VsZXN8MTc0MjEyNTQ5Mw&_rt_nonce=01974a64b3

Nordahl, S. L., Devkota, J. P., Amirebrahimi, J., Smith, S. J., Breunig, H. M., Preble, C. V., Satchwell, A. J., Jin, L., Brown, N. J., Kirchstetter, T. W., Scown, C. D. (2020) Life-cycle greenhouse gas emissions and human health trade-offs of organic waste management strategies. *Environ Sci Technol* **54,** 9200-9 -- https://doi.org/10.1021/acs.est.0c00364

OECD (2022) Plastic pollution is growing relentlessly as waste management and recycling fall short, says OECD. *Last accessed:* 30/08/2023 -- https://www.oecd.org/newsroom/plastic-pollution-is-growing-relentlessly-as-waste-management-and-recycling-fall-short.htm

Open Access News (2022a) "Advanced recycling" not the answer to plastic pollution crisis. *Last accessed:* 02 Sep 2023 -- https://www.openaccessgovernment.org/advanced-recycling-plastic-pollution-crisis-industry-environment/136176/

Open Access News (2022b) The UK to end plastic pollution by 2040. *Last accessed:* 02 Sep 2023 -- https://www.openaccessgovernment.org/the-uk-to-end-plastic-pollution-by-2040/146836/

Ow, C., Taylor-Hearn, I., Wunderley, L., Taylor, S., Pool, M., Fostier, M. (2025) Sustainable SDS-PAGE and Western blotting: cutting plastic, not corners. *The Biochemist* **47,** 3-8 -- https://doi.org/10.1042/bio_2025_106

Pandey, P., Dhiman, M., Kansal, A., Subudhi, S. P. (2023) Plastic waste management for sustainable environment: techniques and approaches. *Waste Dispos Sustain Energy* **5,** 205-22 -- https://doi.org/10.1007/s42768-023-00134-6

Prokop, A. (2018) Why funding fruit fly research is important for the biomedical sciences. *Open Access Govern* **20,** 198-201 -- https://tinyurl.com/y7b25jpmroo

Ragazzi, I., Farley, M., Jeffery, K., Butnar, I. (2023) Using life cycle assessments to guide reduction in the carbon footprint of single-use lab consumables. *PLoS Sustain Transform* **2,** e0000080 -- https://doi.org/10.1371/journal.pstr.0000080

Rizan, C., Bhutta, M. F., Reed, M., Lillywhite, R. (2021) The carbon footprint of waste streams in a UK hospital. *J Clean Prod* **286,** 125446 -- https://doi.org/10.1016/j.jclepro.2020.125446

Roote, J., Prokop, A. (2013) How to design a genetic mating scheme: a basic training package for Drosophila genetics. *G3 (Bethesda)* **3,** 353-8 -- http://doi.org/10.1534/g3.112.004820 http://doi.org/10.1534/g3.112.004820 & http://dx.doi.org/10.6084/m9.figshare.106631

Sever, L., Decorte, M. (2024) From waste to water: Biogases' contribution to a climate-resilient water system. *Blog post in* "European Biogas Association" -- https://www.europeanbiogas.eu/from-waste-to-water-biogases-contribution-to-a-climate-resilient-water-system

Shen, M., Hu, T., Huang, W., Song, B., Qin, M., Yi, H., Zeng, G., Zhang, Y. (2021) Can incineration completely eliminate plastic wastes? An investigation of microplastics and heavy metals in the bottom ash and fly ash from an incineration plant. *Sci Total Environ* **779,** 146528 -- https://doi.org/10.1016/j.scitotenv.2021.146528

Smith, W. V., Bebbington, A., Sircar, R., Pulver, S. R. (2025) Doctoral students as carbon accountants: calculating carbon costs of a PhD in neuroscience. *bioRxiv*, 2025.01.20.633775 -- https://doi.org/10.1101/2025.01.20.633775

Stocker, H., Gallant, P. (2008) Getting started: an overview on raising and handling *Drosophila*. *Drosophila. Methods and Protocols* **420,** 27-44 -- https://doi.org/10.1007/978-1-59745-583-1_2

Taylor-Hearn, I. (2023) Top tips from Gold LEAF champions. *Blog post in* "BMH Social Responsibility" -- https://blogs.manchester.ac.uk/bmh-sr/2023/09/18/leaf-tours/

ThermoFisher, Plastic Materials Selection. *Last accessed:* 04 Sep 2023 -- https://www.thermofisher.com/uk/en/home/life-science/lab-plasticware-supplies/plastic-material-selection.html

Trusler, E. C., Davies, M., Spurrier, B., Gould, S. J. (2024) Reusable glassware for routine cell culture—a sterile, sustainable and affordable alternative to single-use plastics. *Front Sustain* **5** -- https://doi.org/10.3389/frsus.2024.1447236

UK Government (2021) Environment Act 2021. -- https://www.legislation.gov.uk/ukpga/2021/30/contents

Urbina, M. A., Watts, A. J. R., Reardon, E. E. (2015) Labs should cut plastic waste too. *Nature* **528,** 479 -- https://doi.org/10.1038/528479c

Uy, B., Read, H., van de Pas, S., Marnane, R., Casu, F., Swift, S., Wiles, S. (2022) The efficacy of commercial decontamination agents differs between standardised test settings and research laboratory usage for a variety of bacterial species. *PeerJ*, 10:e13646 -- https://doi.org/10.7717/peerj.13646

Wattam, K. J., Todd, M. A. M., Groth, A. (2024) Implementing sustainable science in the lab. *Nat Cell Biol* **26,** 2-3 -- https://doi.org/10.1038/s41556-023-01303-9

Weber, P. M., Michelsen, C., Kerou, M. (2025) What's in our bin? *EMBO Rep* **26,** 297-302 -- https://doi.org/10.1038/s44319-024-00360-x

Wellcome Trust (2024) Concordat for the Environmental Sustainability of Research and Innovation Practice. *Last accessed:* -- https://wellcome.org/what-we-do/our-work/environmental-sustainability-concordat

Wojnowska-Baryła, I., Bernat, K., Zaborowska, M. (2022) Plastic waste degradation in landfill conditions: the problem with microplastics, and their direct and indirect environmental effects. *Int J Environ Res Public Health* **19,** 13223 -- https://doi.org/10.3390/ijerph192013223

Yang, Z., Lü, F., Zhang, H., Wang, W., Shao, L., Ye, J., He, P. (2021) Is incineration the terminator of plastics and microplastics? *J Hazard Mater* **401,** 123429 -- https://doi.org/10.1016/j.jhazmat.2020.123429

Zhao, L., Zhang, F.-S., Chen, M., Liu, Z., Wu, D. B. J. (2010) Typical pollutants in bottom ashes from a typical medical waste incinerator. *J Hazard Mater* **173,** 181-5 -- https://doi.org/10.1016/j.jhazmat.2009.08.066

Zikhathile, T., Atagana, H., Bwapwa, J., Sawtell, D. (2022) A review of the impact that healthcare risk waste treatment technologies have on the environment. *Int J Environ Res Public Health* **19,** 11967 -- https://doi.org/10.3390/ijerph191911967

Figures and Tables

Fig.1. Overview of current waste production and management at the Manchester Fly Facility. **A)** 18°C walk-in incubator at the Manchester Fly Facility holding trays with thousands of individual fly stocks. **B)** Tray with ready-to-use food-filled fly vials sealed in a plastic bag. **C)** Fresh versus used polystyrene vials and polypropylene bottles, closed with a celluloid plug (left vial), cotton wool (right vial) or high density/cellulose acetate plugs (bottles). **D)** Fly waste in plastic bags kept in an upright freezer at -20°C for 48h. **E)** Clinical waste incineration bags holding one or two bags of fly waste. **F)** Pictogram of an incineration facility followed by data describing waste produced and costs arising over a year; total costs include those for freezing and incineration. For underlying data calculations see Suppl. Mat. 1.

Fig.2 Calculated costs for different container types and waste/resource management procedures in single and re-use scenarios over 10 years. Single-use options are shown in **A**, re-use options in **B**. Calculations for container materials (blue) include the production of 1.2 million vials and 20,000 bottles in A and 66,000 vials and 1,100 bottles in B, in both cases shown for different materials used (PP, polypropylene; PS, polystyrene). The GMO-treatment procedures can be freezing (light yellow) or autoclaving (orange). In the re-use scenario, the containers may be washed by hand or in a washer/dryer (light orange) and autoclaved after each wash (5 runs/month) or less frequently (2 runs/month; both darker orange). The amount of waste generated is 4 tonnes in A and 3.2 tonnes in B (after separation from the containers). In both A and B, the freezing step occurs before the containers are cleaned, so 4 tonnes of waste are frozen. Waste disposal procedures (green) are calculated for 4 tonnes in A and 3.2 tonnes in B. For each item, the top bar indicates environmental cost in kg CO_2e, and the bottom bar (hatched white) the economic cost in British pounds. For underlying data calculations see Suppl. Mat. 1.

Fig. 3. Images showing the step-by-step protocol for washing and reusing plastic (Polypropylene) fly vials and bottles in the Raff lab at the Sir William Dunn School of Pathology, University of Oxford. **A)** Used and frozen fly vials and bottles with celluloid plugs in a large box. **B)** Used fly vials and bottles soaked in tap water for at least 2 hours without celluloid plugs in a large box. **C)** The food waste from fly vials and bottles scraped off with a spatula. **D)** Fly waste strained through a sieve. **E)** Fly waste and celluloid plugs discarded for further autoclaving and incineration. **F)** Examples of fly vials and bottles without the fly food. **G)** Fly vials and bottles washed in 1% Chemgene solution with a bottle brush. **H)** Fly vials and bottles soaked in 1% Chemgene solution. **I)** Fly vials and bottles washed with tap water and rinsed with distilled water. **J)** Air-dried fly vials and bottles in a box. **K)** Labels are rubbed off containers with 100% ethanol. **L)** Example of a fly vial autoclaved once (1X) and twenty times (20X); bottles undergo a similar process (not shown). **M)** Example of fly vials in a tray after autoclaving. Note that the plastic trays are non-autoclavable and were soaked in 1% bleach for 10-15 minutes, then rinsed with tap water followed by distilled water. Bottles were kept in fresh cardboard (FLY1016) or fully autoclavable aluminium (FLY1164) trays for re-use (not shown).

Fig 4. Overview of the advantages and disadvantages of the various material choices and options for waste and resource management. In each column, we present the options ranked by expected degree of wastefulness where each tile contains further information relevant for the ranking and to be considered; the colour code indicates whether the option can be used for single-use, re-use or both (see bottom-left inset).

Fig.5 Comparing different combinations of plastic and waste management procedures. Data and colour codes are the same as in Fig.2 and consider the single-use of 1.2 million vials and 20,000 bottles over a period of 10 years, or the re-use of 66,000 vials and 1,100 bottles over 10 years. Different plastics and waste management procedures are colour-coded as indicated in the inset. Four hypothetical scenarios are shown (two single and two re-use), each with different combinations of waste and resource management procedures. In each scenario, the left column shows the environmental emissions in kg CO_2e, the right column (hatched white) indicates the economic costs in pounds. For underlying data calculations see Suppl. Mat. 1.

		supplier-derived information				Calculated	
Container	Supplier	Supplier's reference number	diameter [mm] x height [mm]	Weight g/unit	Cost per unit £	Production CO2eq g /g	CO_2e g/unit
Vial PS	Regina Industries LTD	P1068/T	25 x 95	6.02	0.09	3.8 (1)	22.9
Vial PP	Fisher Scientific	15820275	29 x 95	6.05	0.15	2.5 (1)	15.1
	Scientific Laboratory Supplies / Flystuff	FLY1318/32-120			0.11		
Vial glass	Regina Industries LTD	FB10024	24 x 100	18	0.30	1.4 (2)	25.2
Bottle PP	Scientific Laboratory Supplies	Flystuff 8oz Round Bottom FLY1012	43 x 132	17.66	0.99	2.5 (1)	44.2
Bottle glass	Scientific Laboratory Supplies	Flystuff Half Pint FLY1086	63 x 136	245	7.48	1.4 (2)	343.0

Tab 1. Origin, dimensions, estimated production-related carbon footprint (*cradle to gate*) and purchase cost of fly containers used at or available to the Manchester Fly Facility. Information in columns 3-6 was provided by the suppliers listed and hyperlinked in column 2. For underlying calculations of columns 7 and 8 see Methods. References: **(1)** (Ragazzi et al., 2023), **(2)** (Departments for Energy Security and Net Zero and for Business Energy & Industrial Strategy, 2023). For comparison, the Oxford laboratory used the following containers: PP vials, FLY1318 (£0.31 per vial); PP bottles, FLY1193 (£0.72 per bottle); plastic trays, FLY1090 (£30.75 per tray).

	GMO compliance treatment		Disposal		
	Freeze 48 hrs @ -20°C	Autoclave 2 min @ >146°C	Landfill (household residual)	Incineration lowT (850°C) with EfW*	Incineration highT (1000°C) with EfW
Emissions in [CO_2eq kg / tonne of waste]	73	270	500	175	785
Cost / tonne of waste [£]	76	75	160	270	630

Tab. 2. Estimation of the carbon footprint and cost per tonne of disposed material (*use to grave*). Data were calculated as detailed in the Methods section (EfW, energy recovery).

Appliance	Required use	Consumption	Annual consumption [kWh]	Annual emission [kg CO_2e]
-20°C Freezers	2 freezers @ 24/7	2 x 2 kWh/day	1460	292
Washer (95°C) and dryer (85°C)	25 runs / month	4.5 kWh/run	1350	270
Autoclave at T >121°C for 30 min after each wash	5 runs / month	22.5 kWh/run	1350	270
Autoclave T >121°C for 30 min only when needed	2 runs / month	22.5 kWh/run	540	108

Tab 3. Required use and resultant carbon footprints for appliances involved in re-use procedures. Underlying calculations for the emissions are explained in the Methods section.

Institution	Container materials	Re-use?	GMO compliance treatment	Disposal method
Manchester, UK	PS vials, PP bottles	no	freeze	highT
Bloomington, US*	glass	yes	autoclave (just to kill flies and soften food for cleaning)	rayon plugs sent to landfill; food and flies disposed into sanitary sewer and water treatment plant
Natl. Ctr. Biol. Sci., Bangalore, India	glass vials, PP bottles	yes	autoclave	cotton plugs incinerated at highT; food and flies sent to in-house sewage treatment plant
Univ. Oxford (Raff lab), UK**	PS vials and bottles	yes	freeze/autoclave	cotton plugs sent to highT incineration; discarded containers sent to recycling firm; food and flies sent to incineration
Univ. Cambridge, UK	plastic vials and bottles	yes	autoclave	all is sent to landfill
Univ. Bournemouth, UK	PP vials and bottles	both (40 % re-used)	freeze	highT Incineration
Univ. Oxford (Ctr. Neur. Circuits & Behav.), UK	PS vials and bottles	no	freeze	lowT Incineration
Crick institute, London, UK	PS vials and PE bottles	no	freeze	incineration
Imperial College London, UK	PS vials and PP bottles	no	freeze	lowT incineration

Tab. 4. Overview of waste and resource management practices at Manchester and eight other fly facilities. Columns list the location of fly facilities, the materials of containers used, whether containers are single- or re-used, what GMO compliance measures are taken and the disposal method. Comments: * more information provided in Box 2; ** more info in Box 1.

Figure 1
188x101 mm (x DPI)

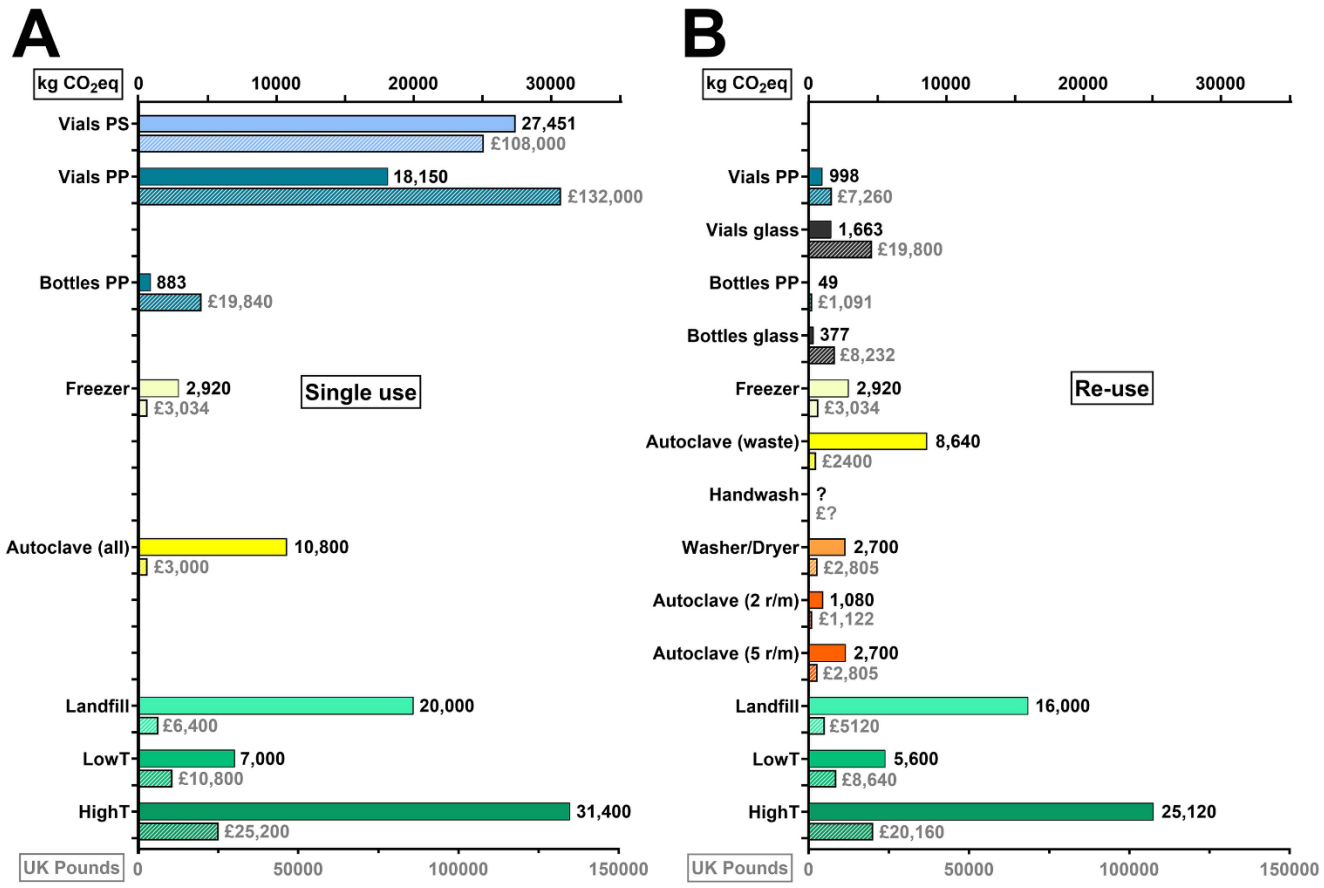

Figure 2
250x169 mm (x DPI)

Figure 3
359x337 mm (x DPI)

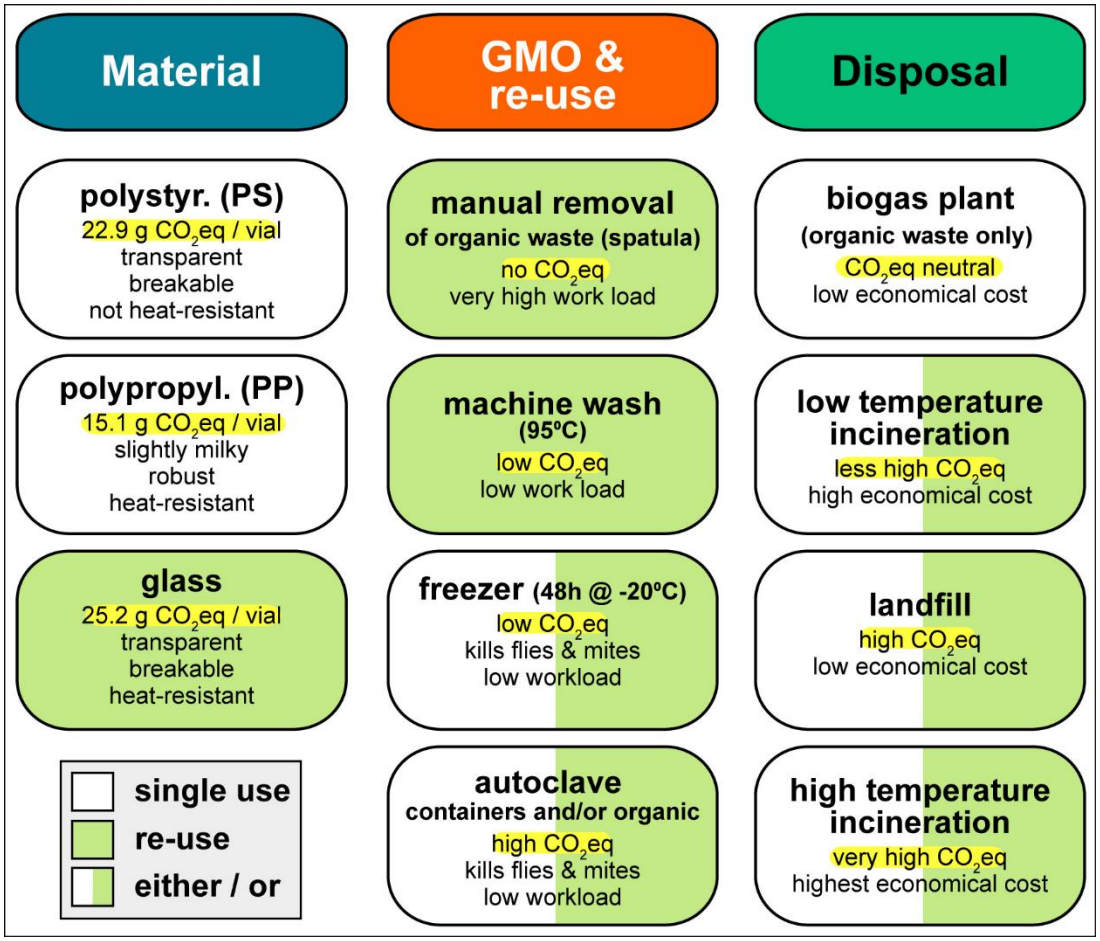

Figure 4
144x123 mm (x DPI)

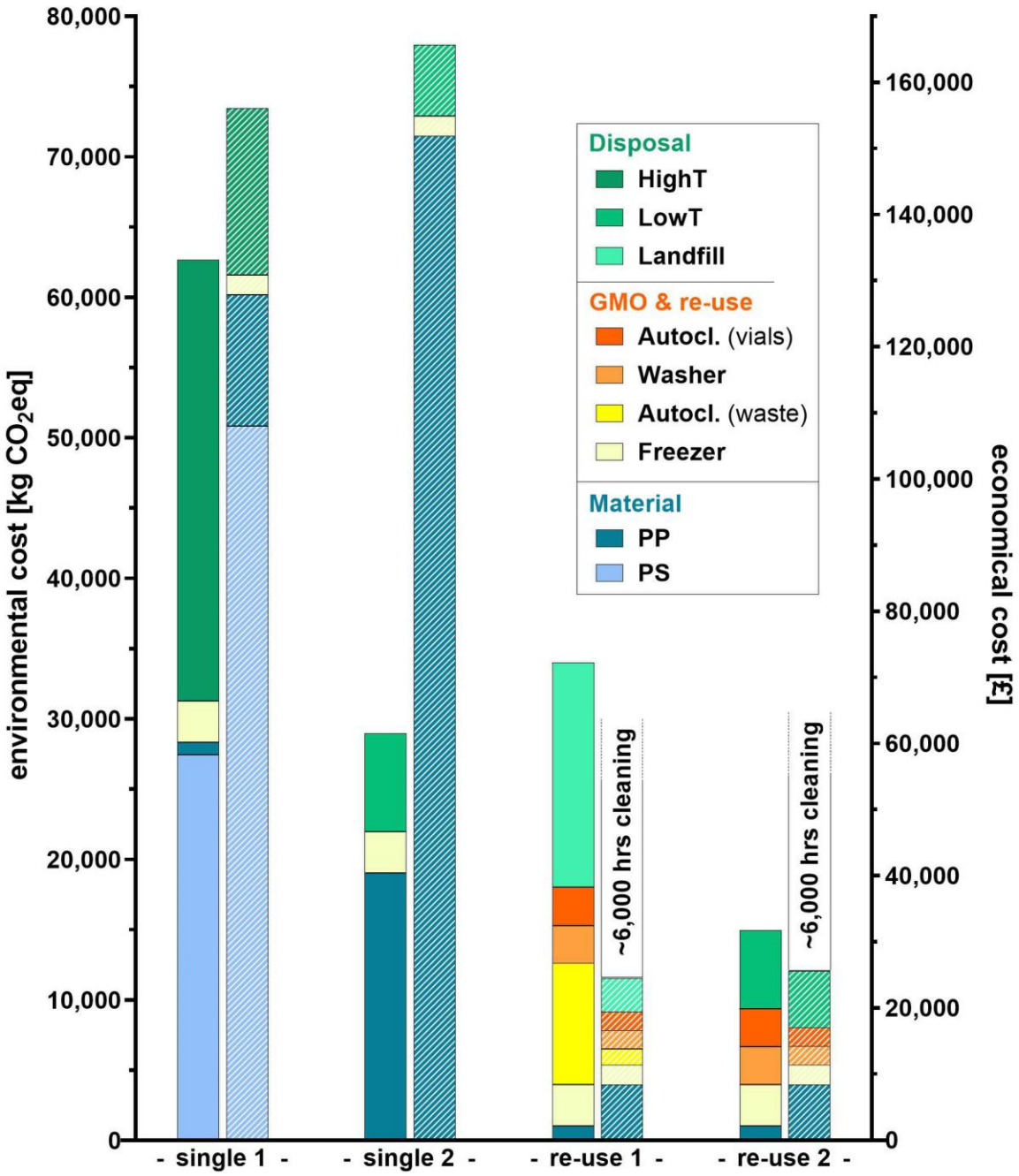

Figure 5
165x190 mm (x DPI)