Nutrient and moisture transfer to insect consumers and soil during vertebrate decomposition

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Abstract

Decomposition of organic matter leads to the redistribution of nutrients to organisms and the environment. Yet knowledge of this process has focused largely on plant-derived organic matter, with little known about relative quantities of nutrients and moisture transferred from decomposing animal remains to insect consumers and soil. We used a replicated and spatially blocked experiment to quantify the moisture, carbon, nitrogen, and phosphorous content of rabbit carcasses, maggot consumers, and soil over 20 days of decomposition.

We found that maggot biomass reached 22% of the fresh rabbit carcass, or 39% of the consumable soft tissues. Maggots were comprised of 68% moisture, and their dry mass was comprised of 25% carbon, 4.9% nitrogen, and 0.8% phosphorous. Soils accumulated approximately 12.9% of the total carcass moisture, but only 0.7% of the carcass dry mass. The largest quantity of carcass mass loss was attributable to evaporation of moisture to the atmosphere (45%). Approximately 9% of the initial carcass mass was left as unconsumed remains. Our study provides estimates of the quantities of nutrients moving from vertebrate carcasses to insect consumers and soil. This knowledge is critical to scaling up the effects of carcasses and to developing our understanding of their role in biogeochemical cycling in ecosystems.

Keywords: carrion, decay, Diptera, nutrient cycle, trophic

1. Introduction

The decomposition of dead organic matter is central to the functioning of all ecosystems and has a major role in the redistribution of nutrients and energy (Swift et al. 1979; Benbow et al. 2018). Dead organic matter comes in many forms, including both plant and animal tissues, and pathways of mass loss and nutrient flow from dead matter to consumers and the broader environment can vary. For example, distinct communities of arthropods, including isopods and millipedes, are involved with the breakdown of leaf litter (Hattenschwiler et al. 2005), whereas flies are the principle arthropod consumers of animal carrion (Payne 1965). Yet, compared with that of dead plants, knowledge of the contribution of dead animals to nutrient cycling is poorly understood, or downplayed (Moore et al. 2004; Barton et al. 2013; Benbow et al. 2018). In particular, studies of the absolute quantities of key macronutrients transferred from dead animals to consumers are rare, and this severely constrains our understanding of the role of carrion in ecosystems (Wilson and Wolkovich 2011; Barton et al. 2013; Subalusky et al. 2017).

Vertebrate carcass decomposition creates a localised island of intense biological activity (Carter et al. 2007). Indeed, large vertebrate carcasses arguably host some of the most species-dense communities of organisms in nature. This is largely due to the high concentrations of nutrient-rich tissues that comprise a vertebrate, and the fierce competition associate with its consumption. This leads to the rapid dispersion of carrion nutrients into local food webs, and the broader environment. There is a good understanding of the diversity of consumers of carrion, and their relationship to other trophic groups (Braack 1987; Wilson and Wolkovich 2011), but knowledge of the quantity of carrion-derived nutrients that transfers to consumers remains poor. Further, there are now multiple studies demonstrating the changes in soil nutrients beneath decomposing vertebrate carcasses from both ecological

(Parmenter and MacMahon 2009; Macdonald et al. 2014; Barton et al. 2016) and forensic perspectives (Benninger et al. 2008; Finley et al. 2015; Singh et al. 2018), and these have shown notable inputs of key macronutrients or changes to soil biotic communities.

Quantitative field studies of nutrient flow from decomposing carcasses through multiple pathways into organisms and the environment are rare (Subalusky et al. 2017). Two examples researching the energy content (Putman 1978a) and CO_2 output (Putman 1978b) of decomposing vertebrate carcasses are the only examples known to us that provide estimates of total nutrient quantities. Further, there have been no field studies conducted to quantify the relative amounts of nutrient transfer from carcasses into both insect consumer and soil recipients. This leaves a significant gap in our knowledge of the rates and quantities of nutrient movement from vertebrate carcasses, and the role of insect consumers in this process. This is important because there is increasing interest in quantifying the potential role of carcasses in contributing to biogeochemical cycling in ecosystems (Parmenter and MacMahon 2009; Barton et al. 2013), as well as the role of insects in providing ecosystem services associated with carrion removal (Tomberlin et al. 2017).

Here we report on a replicated and spatially blocked experiment designed to quantify the effect of time on vertebrate carcass mass change as a consequence of consumption by fly larvae and fluid leakage into soil. We conceptualized mass change as occurring through four main pathways, including assimilation into fly larvae biomass, evaporation and production of gases, scavenging by vertebrates, and movement of fluids into soil (Figure 1). This figure is simplified to ignore the numerous higher trophic interactions occurring at carcasses, such as the predator and parasitoid arthropods (Braack 1987), and we regarded any consumption of the carcass by ants or beetles, as small compared to the action of flies (Payne 1965; Barton and Evans 2017). We also deliberately excluded vertebrates to reduce further the complexity of possible consumers, and did not directly measure gas production or evaporation. This

allowed us to use the equation: initial carcass mass = maggot mass + soil mass + remains. We quantified both wet and dry mass of each of these three components, and further partitioned dry mass into carbon, nitrogen and phosphorous components. Our study provides, for the first time, a 'per-carcass' estimate of nutrient and moisture transfer to insect consumers and soil under particular conditions, thus allowing for the scaling of carrion input to insect food webs and ecosystem biogeochemical cycling.

2. Methods

2.1 Study area and experimental design

We conducted our study in Goorooyarroo Nature Reserve near Canberra, south-eastern Australia (Shorthouse et al. 2012). We selected a five-hectare area of grassland that was dominated by the native perennial grass *Themeda australis*, and had a silty-loam soil with A-horizon to a depth of approximately 10-15 cm. We used a randomised block design of 25 carcasses grouped into five blocks of five carcasses each across the study area. Carcasses were of introduced wild European rabbits (*Oryctolagus cuniculus*) killed by firearms during routine pest control operations. All rabbits were killed on the same night, and had a similar time of death. Only intact and mature adult rabbits were used so as to minimize the variation in size and quality of the carcasses. All carcasses were placed into refrigeration on the same night, and remained refrigerated at 3ºC for 48 hours until deployment in the field. All individual carcasses were weighed and their initial starting mass recorded. We paired each carcass with a control site without a carcass, 1-metre away for use in deriving soil nutrient differences, and thus carcass inputs. We determined that a distance of 1-metre between each carcass and its control site as was sufficient to prevent contamination while also minimizing local differences in soil properties due to natural heterogeneity. We ensured that each carcass

was placed on level ground so as to avoid potential movement of fluids and contamination of control sites. Experimental blocks were approximately 40 metres apart, and carcasses within each block were spaced approximately 5 metres from each other. We deployed all carcasses during the morning of the first day of the experiment (day 0). We collected a single carcass at random from each block (n=5) every four days, thus providing data at days 4, 8, 12, 16, and 20. Previous empirical data has shown this amount of time is sufficient to capture the majority of mass loss under similar abiotic conditions (Barton and Evans 2017).

We collected temperature and rainfall data using an on-site rain gauge and temperature data loggers (TC Thermochron®). We placed a temperature logger under three different rabbit carcasses and their paired control sites. We programmed the loggers to collect temperature data every 30 minutes for the entire duration of the experiment. We summed data for every 24 hr period, then divided by 48 to give a mean daily temperature. We then summed mean daily temperatures to give accumulated degree days (ADD) (Megyesi et al. 2005), which is a biologically meaningful integration of time and thermal energy underpinning insect development and other metabolic processes.

2.2 Field sampling protocol

We collected carcasses and soil samples at days 4, 8, 12, 16, 20. We lifted the carcasses off the ground and placed them into plastic bags (Fig. 2a), including all remains and maggots, and transported them to the lab where they were temporarily stored in a refrigerator to halt decomposition and maggot activity until laboratory processing. We collected additional visible maggots from the ground under the carcass and placed those in separate containers (Fig. 2b). We gave careful attention to ensure that we obtained a complete sample of all carcass remains and maggots. We did not separate out larval instars or attempt to identify all species, and thus could not estimate demographic change or accurate population sizes.

However, visual observations confirmed that hairy maggots (*Chrysomya* spp.) were the most abundant in the maggot masses, as shown by previous research in this area (Barton and Evans 2017). Soil samples were taken at each carcass and paired control site using cores (50 mm diameter) to a depth of 30mm directly beneath each carcass (Fig. 2c). Three soil cores were taken for nutrient analysis, pooled on site, and then transported to the laboratory where they were allowed to air dry for several days. We took an additional core using a bulk density ring to a depth of 30 mm for determination of soil density and moisture content. These samples were sealed in the field to prevent moisture loss, then transported to the laboratory. Finally, we measured the length and width of the soil surface area covered by each carcass after they were lifted from the ground. We treated the surface area under each carcass as a proxy for the effect of each carcass on the soil, and multiplied soil core nutrient concentrations by area to determination total soil inputs.

2.2 Laboratory sample processing

After refrigeration at 3 °C (<48 hours), maggots and attached plant debris were removed from carcasses. Further sorting of maggot samples was performed to remove unwanted plant or soil matter. Fresh mass of both maggots and carcass remains were measured using calibrated balances (A&D FA2000, ± 0.01g, or A&D HF300, ± 0.01g). Maggots and carcass remains were then placed in a drying oven at 40 ºC for five days until a constant mass was achieved, then re-weighed to obtain dry mass values. Differences between fresh and dry mass were calculated to give moisture content (g). Dried maggots were subsampled and ground using a coffee bean grinder, then mortar and pestle, prior to analytical assessment of C/N/P content (see below). For nutrient analysis, we used maggot samples from three different carcasses at each time due to low variability between maggot samples (i.e. consistent stoichiometric ratios in larvae tissues).

For soil analyses, we used all soil samples (n=5) from each time. Soil cores taken for moisture and density analysis were weighed, dried at 105 °C, and then weighed again to determine moisture content and soil density (Rayment and Higginson 1992). Soil cores taken for C/N/P analysis were homogenized by passing samples through a 2 mm sieve, light grinding with mortar and pestle to reduce the size of aggregates, and removal of extraneous organic matter such as litter, plant roots or invertebrates.

The assessment of C/N/P content of carcasses at each sample time was not possible due to difficulties in separating dried tissues from each other. Therefore, fresh carcasses (n=3) and carcasses from day 20 (n=3) were separated into four tissue types: skin + fur, skeletal muscle, internal organs, bones. No internal organs or muscle was left at day 20, and only skin + fur and bones were present. Each tissue type was weighed fresh and after drying for several days at 40 °C. Subsamples of the dried tissues were homogenized using a coffee bean grinder, then mortar and pestle, prior to analysis for C/N/P content.

Total carbon (C) and nitrogen (N) were determined with Dumas dry combustion and conductiometric analysis (Vario Max CNS, Elementar, Germany) (Matejovic 1997). Total phosphorus (P) was determined after Kjeldahl digestion at 370°C, followed by colorimetric analysis of phosphorus as orthophosphate using flow injection autoanalysis (FIA)(Lachat Instruments, Milwaukee, Wisconsin, USA) (Diamond 2006).

2.3 Data analysis

We converted all moisture and nutrient values to both percentages and their mass equivalents (g). For moisture, we calculated the difference between wet and dry mass, and expressed this as a percentage of the original mass. For carcass and maggot nutrients, we multiplied the dry mass by the percentage concentration. For soil nutrients, we first calculated the difference between each carcass and control pair at each time point (day 4, 8, 12, 16, 20) to quantify the

amount added to soil by each carcass. We then took the mean value of the % nutrient differences across all five sample times (days 4, 8, 12, 16, 20), and multiplied this value by soil core mass (density (g/cm^3) x volume ($58.875cm^3$)), thus giving an absolute mass (g) per soil core. To scale this up to a per-carcass value, we divided the soil core area into each carcass area (approx. 420-900 cm^2), then multiplied this factor by the per-core mass to give the total mass of nutrients transferred to the soil at that carcass. We used a paired t-test to compare mean daily temperatures at carcass versus control sites. Values presented in the figures are means and their standard error.

3. Results

3.1 Carcass mass loss and tissue composition

Mean daily temperatures ranged between 17.6 – 32.4 °C (mean = 22.9) at carcasses, and 17.9 – 22.2 (mean = 20.4) at control sites (P = 0.003), and accumulated degree days reached 459 at carcasses and 408 at control sites (Figure S1). Total mass loss of rabbit carcasses after 20 days of decomposition was over 90%, with a mean starting wet mass of 1456 ± 32 g per carcass (day0) and a mean end mass of 136 ± 5 g per carcass (day20). The pattern of mass loss followed a typical negative exponential trend (Figure 3), with carcass moisture content dropping rapidly in the first four days, and dry mass changing relatively little over the subsequent 16 days. All soft tissues (muscle, internal organs) were consumed by day 20 (Figure 4a, Figure 4b), and these comprised approximately 821 g (56%) of the total fresh mass of a rabbit carcass. This represents 628 g of moisture, 92.8 g of carbon, and 23.1 g of nitrogen, and 1.8 g of phosphorous available for consumption by maggots. Bone had a nutrient profile distinct from the other tissue types (Figure 4c) due to the higher phosphorous

content, as well as the substantial calcium component (not measured) that made up the dry mass.

3.2 Maggot production and nutrient composition

The main insect consumers were larvae of the fly species *Chrysomya rufifacies* and *Chrysomya varipes* (Calliphoridae). Maggot masses peaked at 322.2 (± 49.3) g of biomass at day four of the experiment (Figure 5a), with all subsequent measures of resident larvae and pupae declining over time. At day four, the moisture content of maggots was 218.7 g, whereas the nutrient composition of the maggots was 25.9 g (25%) of carbon, 5.1 g (4.9%) of nitrogen, and 0.8 g (0.8%) of phosphorous (Fig. 5b).

3.3 Soil moisture and nutrient composition

We found that carcasses delivered approximately 100 g of moisture to the soil within the first four days, and this was maintained for the duration of the experiment (Fig. 6a). Inputs of moisture and nitrogen occurred during the first four days of decomposition, but phosphorous input continued until day 12 (Figure S2). The input of carbon did not differ greatly from zero, and was only positive on day 12 (Figure S2). Averaging across carcasses, the total input of moisture into the soil was approximately 134 g, whereas carbon was 0.8 g, nitrogen was 1.74 g, and phosphorous was 0.49 g per carcass.

3.4 Summary of nutrient and moisture transfer

We summarise in Figure 7 the relative proportions of moisture, carbon, nitrogen, phosphorous, and ‘other’ components comprising a fresh carcass and transferred to maggots, soil, the atmosphere, and left in the carcass remains. We note the large proportion of ‘other’ components comprising oxygen and hydrogen, and other trace elements present in tissues.

236 The unmeasured components include both measurement error and emission of gases and
237 volatiles (e.g. CO_2).
238
239 ## 4. Discussion
240
241 We set out to quantify the change in mass and nutrient content of rabbit carcasses, and the
242 subsequent change in mass and nutrients of maggots and soil. After 20 days of decomposition
243 only 9% of the initial mass of carcasses remained, with the greatest portion of mass
244 transferred to the atmosphere via evaporation of moisture. The next largest component of
245 moisture transfer was to maggots, and then soil. In contrast to moisture, the largest nutrient
246 (C, N, P) component was left in the remains, followed by transfer of nutrients to maggots,
247 and the smallest fraction went to the soil. Our study provides new information about the
248 relative quantities of nutrients and moisture transferred to distinct parts of an ecosystem. This
249 allows for a new appreciation of the role of carrion in supporting insect food webs and
250 broader ecosystem biogeochemical cycling.
251
252 ### *4.1 Mass loss*
253 The largest quantity of carcass mass was transferred to the atmosphere via the evaporation of
254 moisture. We can infer that this was likely the pathway of mass transfer given our
255 experimental design that excluded vertebrates, and measured maggots, soil, and remains. This
256 finding is not surprising given the relatively warm daytime temperatures experienced during
257 the study (up to 32 degrees C). Warm weather can have either facilitative or inhibitory effects
258 carcass decomposition, including speeding the rates of insect development and biochemical
259 processes, as well as drying effects and mummification (Forbes and Carter 2015). In our case,
260 and despite some rainfall occurring prior to day 12, evaporation of carcass moisture was a

large driver of mass loss. Evaporation is likely to have occurred both directly from the carcass, but also indirectly following moisture transfer to maggots and the soil.

The second largest component of mass was transferred to fly larvae, with maggot biomass peaking at 22% of a whole carcass, or 39% of the consumable soft tissues, during day 4 of decomposition. This value is likely an underestimation as we were not able to determine if fly larvae continued to increase in mass between days four and eight, or how other factors such as maggot massing, competition, or larval mortality may have affected net biomass production (Shiao and Yeh 2008; Johnson and Wallman 2014). The larval masses at day four would likely have continued to consume the flesh of carcasses, but we were not able to determine the total transfer of mass to maggots over the entire duration of the experiment due to metabolism and excretion.

Soils accumulated an average of 134 g of moisture, but only 3 g of macronutrients per carcass, and therefore received the smallest amount of mass from the carcass. Approximately 9% of carcass mass was left as unconsumed remains, but this was largely dry mass with little moisture left. Further inputs of carcass remains to the soil would have occurred over the longer term (Barton et al. 2016), and so it is important to highlight that our study shows nutrient transfers within a specific 20-day timeframe.

The rapid mass loss of carrion is a distinguishing feature of the decomposition of animal-derived biomass (Parmenter and MacMahon 2009; Barton et al. 2013). The consumption and recycling of carcass nutrients to organisms and soil is orders of magnitude faster than that for many forms of dead plant biomass, despite there being much smaller quantities than plant biomass in most ecosystems (Parmenter and MacMahon 2009). The rapid turnover of animal carrion means that its contribution to ecosystem nutrient cycling is probably disproportionate compared with plants, but the magnitude of its contribution is still largely unknown for ecosystems worldwide. In fact, knowledge of actual quantities only exist

for specific forms of carrion, such as a suite of vertebrates in a semi-arid ecosystem in the USA (Parmenter and MacMahon 2009), or wildebeest in a river ecosystem in the Serengeti (Subalusky et al. 2017). Knowledge of the turnover of carrion biomass from a range of animal species in different biomass is completely lacking.

4.2 Moisture and nutrient flow pathways

The decomposition of vertebrate carcasses results in the transfer of nutrients to different organisms and the environment. Yet, each part of a carcass is not equally likely to transfer to these different ecosystem sinks, or to transfer at similar rates. For example, we found in our study that the soft tissues of each rabbit carcass were completely gone by the end of the experiment (20 days), but that the bones and fur remained. These remaining tissue types had very little moisture left, but contained a high proportion of the dry mass, and a particularly large proportion of the phosphorous (due to the calcium phosphate in bones). Another notable finding was that the macronutrient content of skin and fur appeared to decline over the course of the experiment, whereas the nutrient content of bone did not. It is not clear if this was due to leaching of nutrients belowground, and/or changes to proteins and release of gaseous compounds, and/or some contamination and dilution of the fur with soil/dust or when it was collected from the field.

The main process delivering nutrients to the soil is direct leakage of carcass fluids (e.g. blood, extra- and intra-cellular fluids). However, additional fluids can be added via excretion of moisture and ammonium from maggots (Chapman 1998), which are initially sourced from the carcass via tissue consumption. Additional carbon and nitrogen are added to soil as pupal casings (puparia), with burrowing into soil also provide pathways for water infiltration. Burrowing activity of other insect larvae, such as the larvae of predatory beetles (e.g. *Saprinus* spp., F. Histeridae), also add soil pores that enhance fluid infiltration and deliver

311 nutrients deeper into the soil. Belowground populations of nematodes and soil mites may also
312 increase in response to the proliferation of bacterial populations under carcasses (Szelecz et
313 al. 2016; Singh et al. 2018), contributing further to the decomposition 'island' effect (Carter
314 et al. 2007). A peculiar finding was that the stoichiometric ratio of soil nutrient inputs
315 differed from that of the rabbit carcass (and the maggots). Specifically, less carbon than
316 expected entered the soil, with nitrogen dominating the soil enrichment. Typically,
317 stoichiometric ratios of soil substrates reflect those of the inputs (Sardans et al. 2012), but this
318 was not the case in our study. This is perhaps due to the soluble forms of nitrogen, such as
319 ammonia, entering the soil more easily as fluids than large organic compounds and fragments
320 of tissues that did not penetrate the soil surface. We also removed large organic fragments
321 from the soil during processing, including pupal casings, which were sources of carbon
322 originating from the carcass. When undisturbed, and considered over a longer time frame, the
323 contribution of carcasses to soil carbon would likely be higher.

324 One of the key challenges in our experiment was the estimation of maggot production
325 from carcasses. We suggest this may be the largest source of error in our study design, and
326 perhaps contributed most to the 'other' unmeasured quantity of dry mass not attributable to
327 soil or the carcass remains. The measures of maggot biomass are likely an underestimate due
328 to the incomplete consumption of soft tissues at day four, and the likely continued growth and
329 development of the maggot populations after day four. Further, the dispersal of maggots
330 away from the carcasses to pupate meant we only collected those maggots inside or
331 immediately adjacent to each carcass at each time point. All up, this meant we had no way to
332 determine the cumulative maggot production from our sampling protocol. However, the rapid
333 consumption and mass loss of carcasses indicate only a single generation of flies was
334 possible, with the emergence of adult *Chrysomya* spp. (F. Calliphoridae) occurring within 10-
335 12 days in a similar study with similar temperatures (Barton et al. 2017). Of course, we also

did not attempt to quantify any mass transfer into the broader insect foodweb, such as carrion beetles (Silphidae) or hide beetles (Trogidae) (Braack 1987), or ants (Formicidae) (Barton et al. 2017), but this mass was likely to be trivial given the numerical dominance of the maggots.

Important questions remain about the interactions between insect and vertebrate scavengers. We deliberately excluded vertebrate scavengers from our carcasses so as to allow more accurate attribution of consumption by maggots. However, in many ecosystems vertebrate scavengers have a major role to play in the consumption of carrion (DeVault et al. 2003; Wilson and Wolkovich 2011), and may consume many smaller animals completely. Even for carcasses of large animals, scavengers will reduce the resources available to flies, and alter the quantities of nutrients moving through different pathways from that carcass. Experiments that manipulate insect and vertebrate access to carcasses, and quantify their interactive effects on mass loss and nutrient flow, would be valuable for building a more complete knowledge of the role in carrion in supporting complex carrion food webs.

4.3 Synthesis and Implications

We set out to investigate where carcass biomass goes during decomposition, and to quantify the relative amounts that flow into the different soil, insect, atmosphere, and remains nutrient pools. By doing this, we hoped to establish a basis for scaling up results to understand the contribution of different carrion sources to different aspects of ecosystem function. The advantages of this can be appreciated by describing scenarios of carrion turnover in ecosystems, for example by multiplying the number of carcasses that enter a defined ecosystem in a given period of time. For example, to quantify the nitrogen input to soil from a population of 1000 rabbits in a 1000 hectare area, and a 50% population turnover rate per year (and ignoring predation), then we can multiply 500 rabbits x 1.74 g nitrogen = 870 g of

nitrogen entering the soil per year. This, of course, is only for one species of vertebrate and does not consider the full community of vertebrates, their different population densities or dynamics, which would yield a considerably higher quantity. Using the same scenario, the amount of nitrogen dispersed away from carcasses as flies might be calculated as 500 rabbits x 5.1 g nitrogen = 2550 g. Other useful numbers include the numbers of flies arising from the approximately 2500 larvae we collected per carcass. This might result in 500 adults, or a total of 250,000 flies from 500 carcasses per year, all able to redistribute their nutrients several kilometers away from each carcass (Norris 1966).

A further implication of our work is that our data suggest a dynamical model of decomposition and mass loss might be achievable. Many of the key processes involved in carrion decomposition is temperature and humidity dependent (Parmenter and MacMahon 2009; Forbes and Carter 2015). For example, our experiment was conducted over 20 days at a mean temperature of 23 ºC, but the relative quantities of nutrient flow via each pathway might change in different seasons or different locations. At cooler temperatures, for example, moisture evaporation might be less, and this might result in a greater portion of the moisture transfer to maggots or to the soil. In other locations, fly communities might be dominated by other species with different development rates or competitive dynamics. This might lead to different amounts of biomass being consumed over similar timeframes, and therefore different quantities being left as remains or entering the soil, for example. Clearly, the development of a general model of mass change during vertebrate carcass decomposition that allows for changes in abiotic parameters is necessary for it to be broadly applicable worldwide. This can only be achieved by further studies quantifying nutrient flow form carcasses in a range of biomes and in different seasons.

In conclusion, we have given estimates of the quantities of nutrients moving from vertebrate carcasses to insect consumers and soil. This knowledge is important to close the

gaps in knowledge of how where carrion biomass is recycled, how much carrion is in landscapes, and to develop more fully our understanding of the role of animal carcasses in supporting food webs and biogeochemical cycling in ecosystems.

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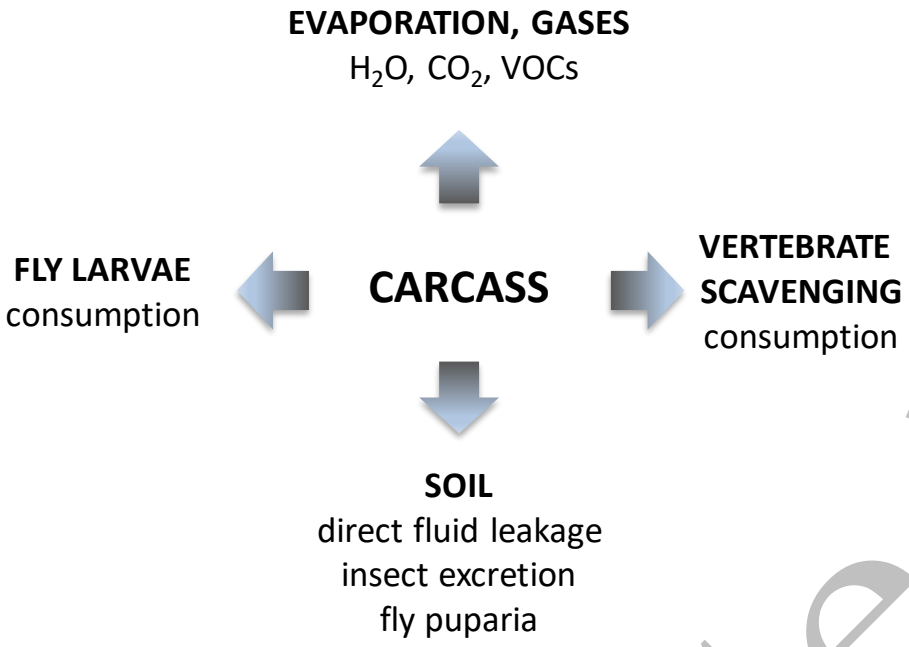

491

492 **Figure 1.** Simplified pathways of nutrient and moisture transfer away from a vertebrate

493 carcass.

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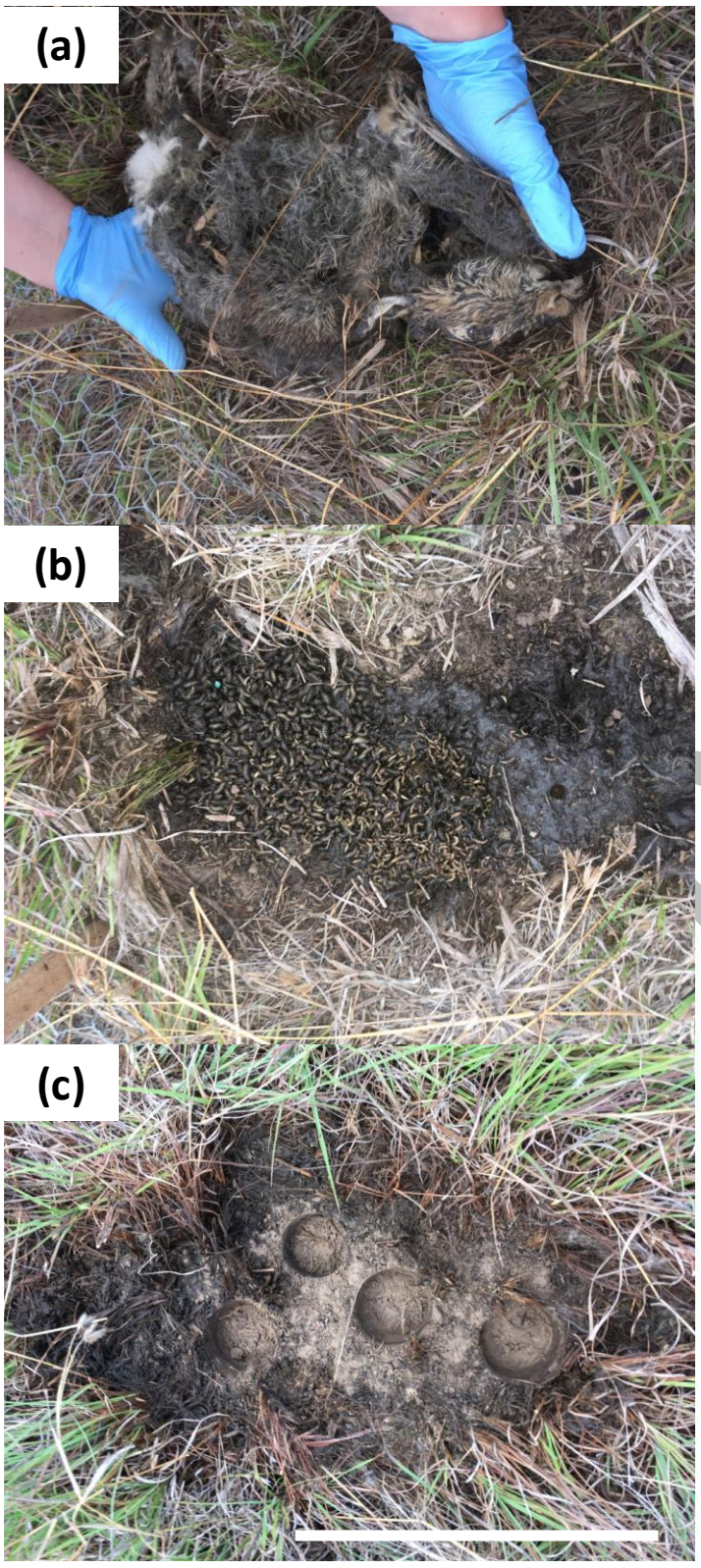


Figure 2. Field sampling protocol followed three steps, including (a) removing the carcass, (b) collection of maggots, and (c) taking soil cores. Samples were processes to obtain total moisture, carbon, nitrogen, and phosphorous content. White bar = 30 cm.

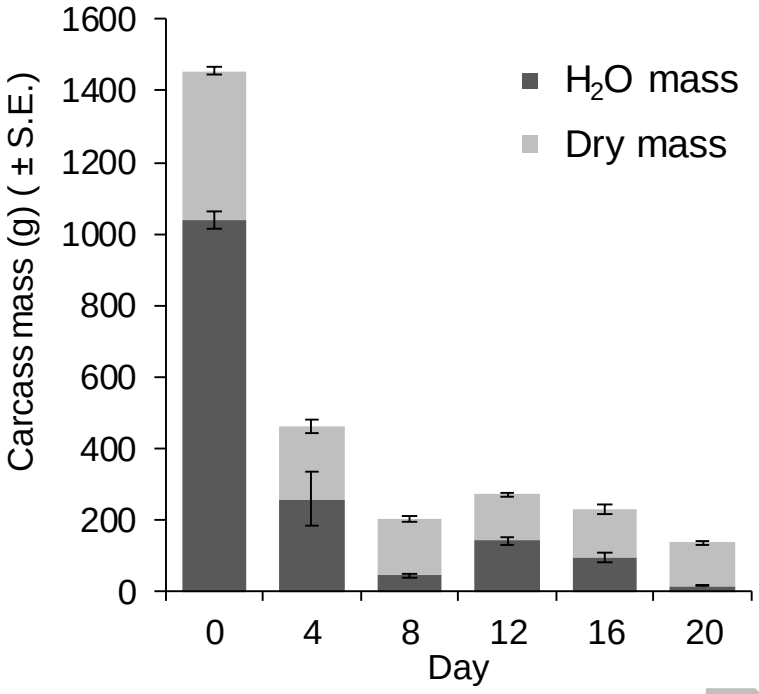
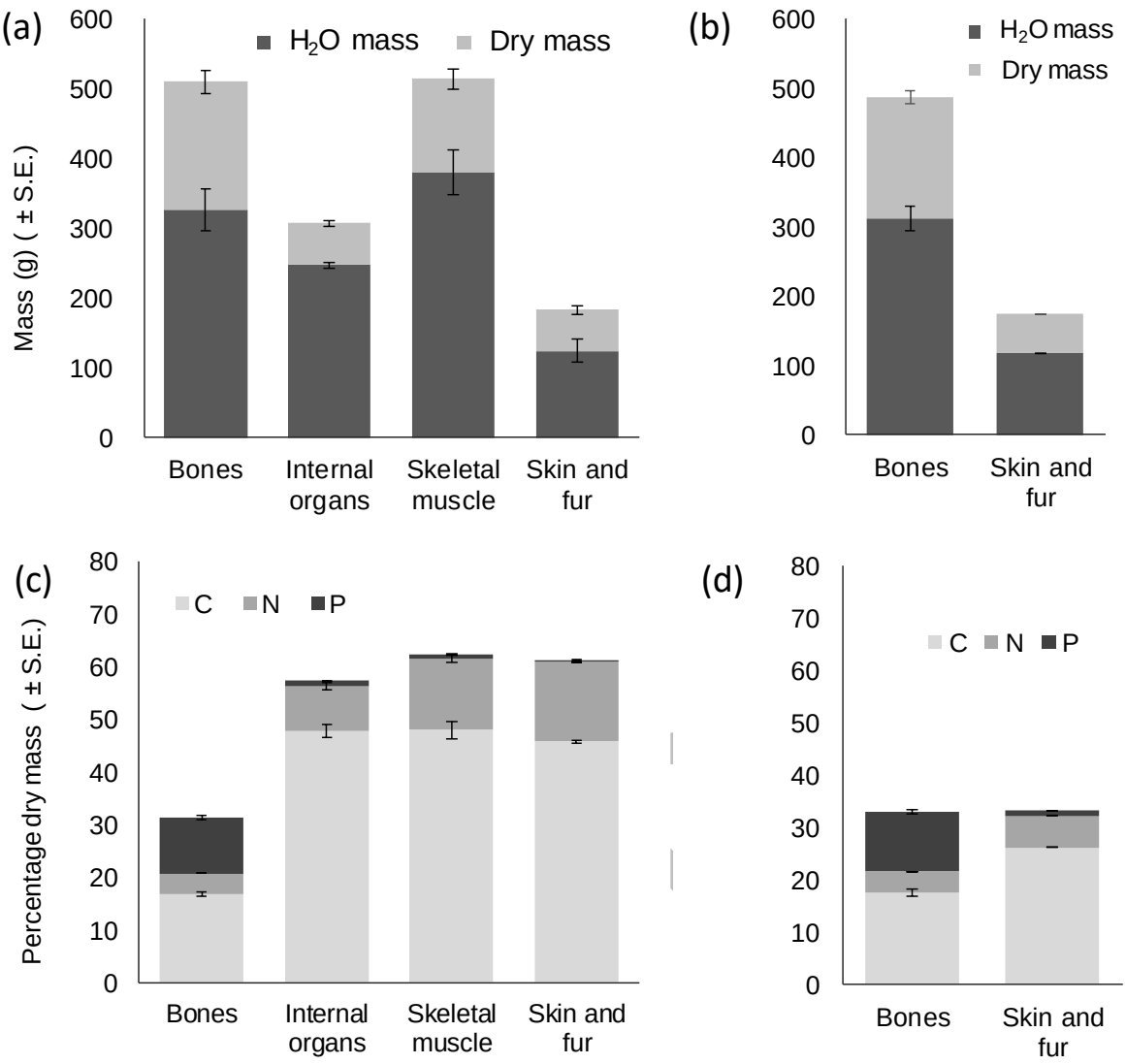


Figure 3. Carcass mass loss followed a negative exponential pattern with moisture loss driving the initial rapid drop in mass. Rainfall occurred after day 8 and this is evident in the small increase in carcass moisture at day 12.



512

513 **Figure 4.** Moisture and nutrient composition of a rabbit carcasses split into four broad tissue

514 types at (a, c) day 0 when carcass was fresh, and (b, d) day 20 when dry remains were left.

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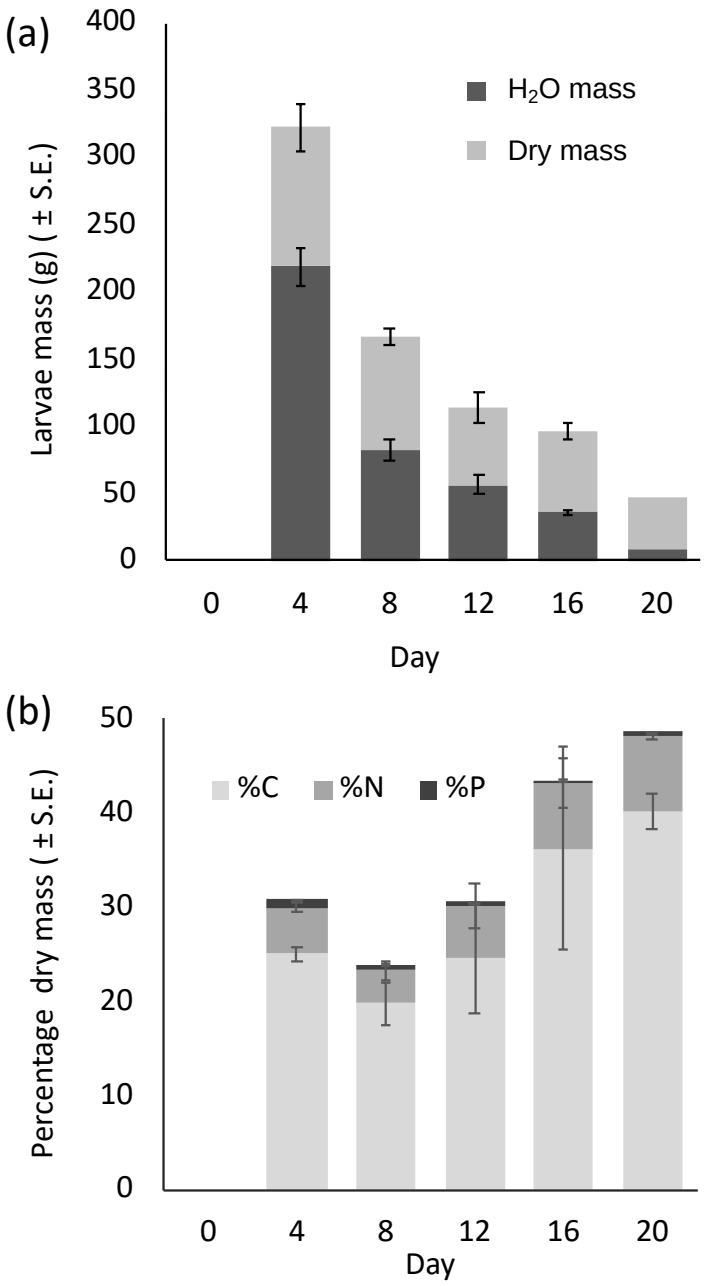

519

520 **Figure 5.** Change in (a) mass and (b) nutrient composition of fly larvae in rabbit carcasses

521 during decomposition.

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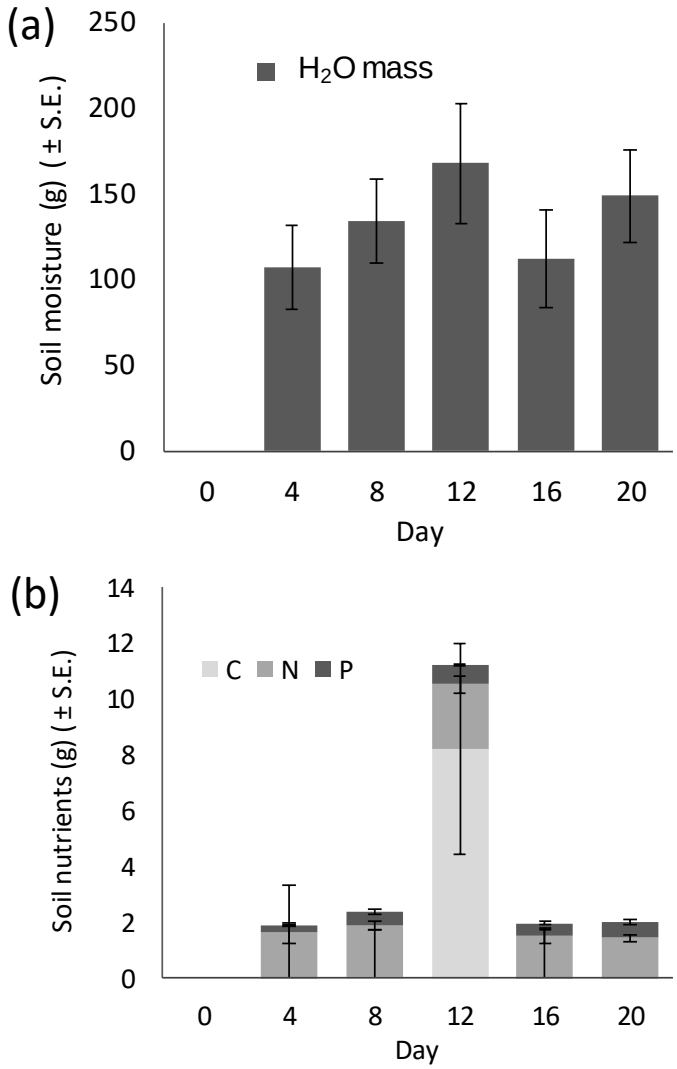

523

524 **Figure 6**. The mean (± s.e.) difference in soil (a) moisture and (b) nutrient content between

525 each of the carcass and control pairs (n=5) every four days of the experiment.

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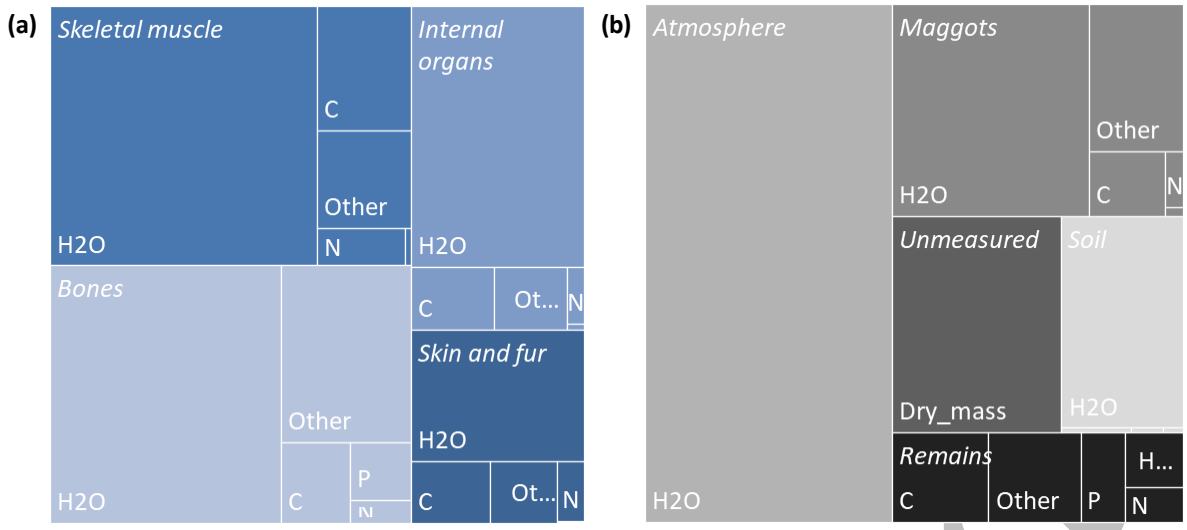

Figure 7. Proportional representation of the moisture and nutrient (C, N, P, other) components of (a) a fresh rabbit carcass, and (b) recipients of the carcass following 20 days of decomposition. Only the soft tissues were able to be consumed by maggots. Moisture loss occurred from all tissues. The atmosphere component includes the left-over moisture not quantified in the maggot, soil, and remains, and includes evaporation/emission of moisture and gases. The unmeasured component includes the dry mass not quantified in the maggot, soil, and remains, and includes likely underestimation of total maggot biomass. All values derived from means, and represent the proportion of the total fresh carcass mass.

539 **Supporting Information**

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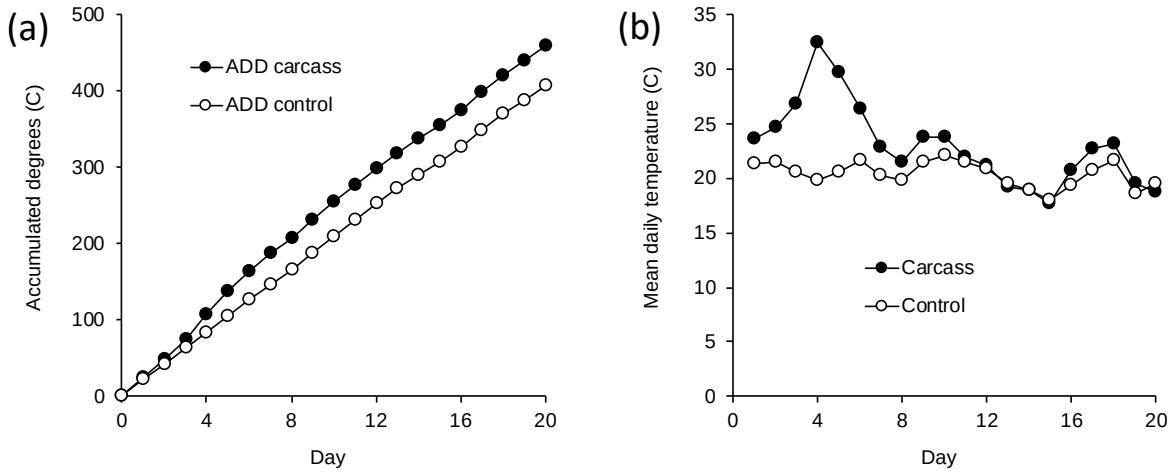

542

543 Figure S1. Differences in temperature were recorded between carcasses and ambient controls.

544 This was evident in (a) a higher number of accumulated degree days at carcasses, and (b)

545 daily temperaatures being 5-10 degrees warmer at carcasses during active decomposition on

546 days 2-6 when maggot masses were most vigorous.

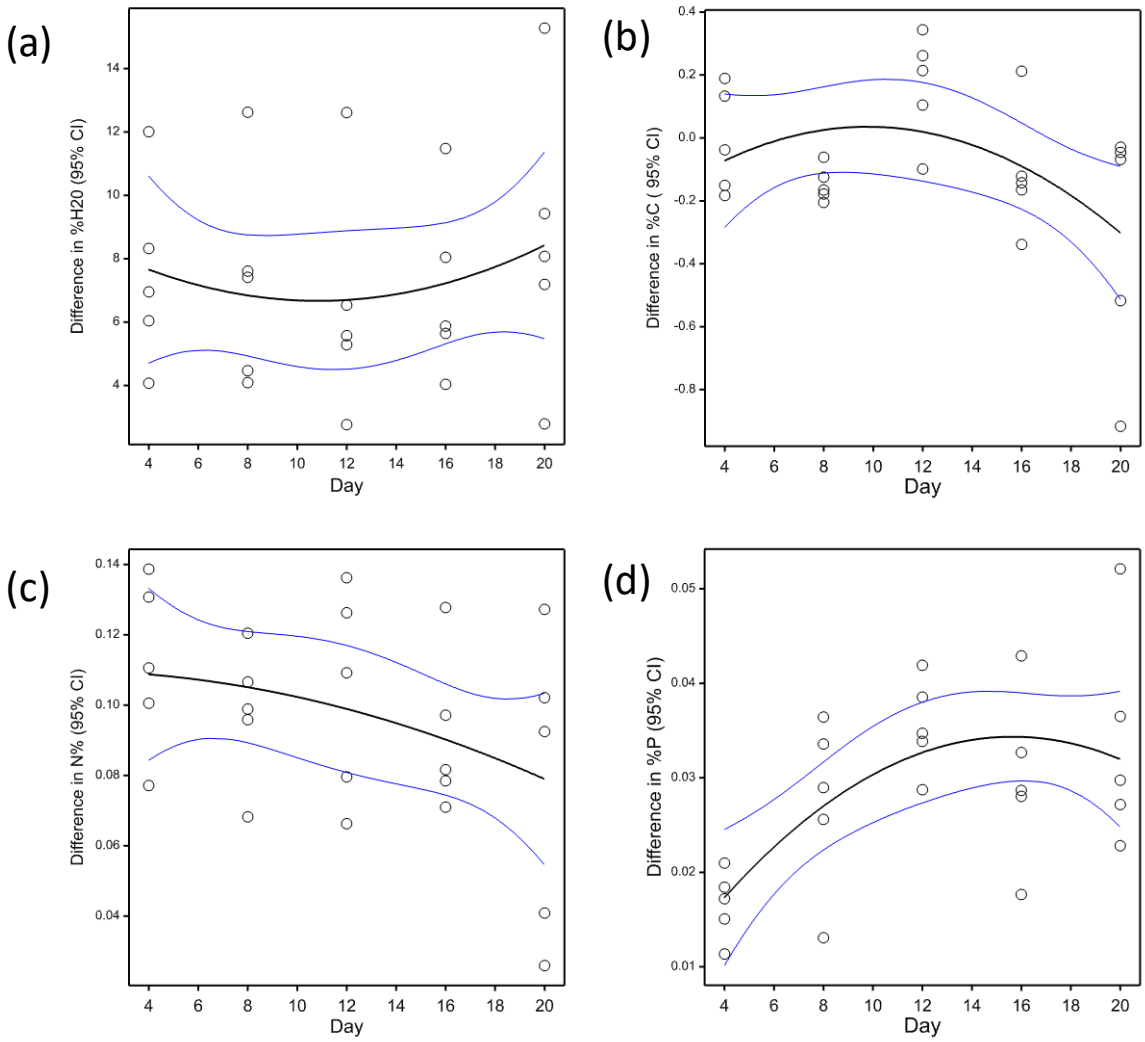

Figure S1. Polynomial regression with 95% CI fitted for differences in (a) H20, (b) carbon, (c) nitrogen, and (d) phosphorous between carcass-control pairs over days 4 – 20 of the experiment.