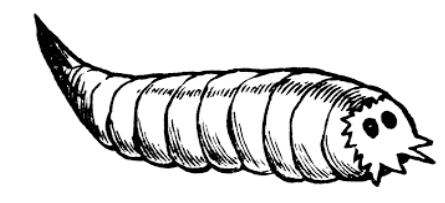




The Effect of Plastic Bags on Decomposition and Necrophagous Fly Composition

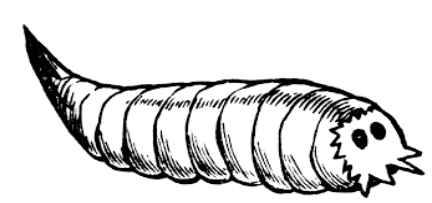
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Introduction

Forensic entomology uses insects to help estimate the post-mortem interval (PMI) based on blow fly colonization (Haskell and Williams 2008). The PMI provides an estimate of the time between death and corpse discovery, and is of extreme importance due to its role in forensic investigations. Many factors can influence the PMI such as temperature, weather, body mass, and insect activity (Haskell and Williams 2008). Insects that typically aid in decomposition are flies, beetles, bacteria, and fungi. The most important insects for PMI estimations are from the order Diptera. These flies are used to estimate the PMI because they are early colonizers with well studied life histories and development times.

Accurate PMI estimations are crucial to forensic investigations, and any factors that influence blow fly colonization are important for entomologists to consider. This study focused on placing plastic bags of different thicknesses over the head of pigs. Flies typically lay their eggs in the mucous membranes (Haskell and Williams 2008), and researchers wanted to see the impact of blocking their preferred oviposition sites. Asphyxia via plastic bag has been documented in case studies in the literature as a means of suicide, homicide and accidental death (Johnstone *et al.* 1960, Haddax *et al.* 1996, Saint-Martin *et al.* 2009). Researchers were interested in the effect of the plastic bags on decomposition and Dipteran oviposition behavior.

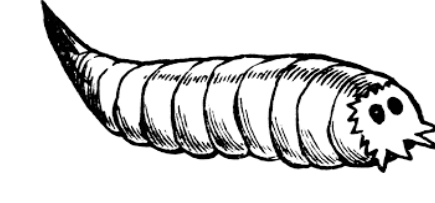


Research Questions

- Does having a plastic bag around preferred oviposition sites impact oviposition timing?
- Does the thickness of the bag have an impact on oviposition timing or subsequent life events?
- Does the presence of a plastic bag influence the Dipteran species composition?



Figure 1. "Team Maggot" Researchers



Methods

Nine fetal pigs were thawed from frozen 24 hours before the start of the experiment. Six fetal pigs were covered with plastic bags: three with thin plastic bags (average bag weight of 5.43 g) and three with thick plastic bags (average bag weight of 11.9 g) (Fig 3-4). The remaining three pigs served as the control group. These pigs were placed outside in a cage (Fig 5), monitored for 6 days, and checked three times daily to look for the presence of blow fly eggs, adults, and maggots. Two trials were done, one commenced on September 27, 2021 and the second on October 19, 2021.



Figure 2. Student researchers with pigs prior to field placement

Data were collected on dates and times that adult blow flies, eggs, first, second, and third instars were found on each pig. Third instar maggots were collected off of each pig with the date of collection recorded, pig number, and type of plastic bag (thick, thin, or none). Data analysis included species identification of each maggot collected and statistical analysis of stages found on the pigs. Species identification was performed on third instar larvae using taxonomic keys derived from (Stojanovich and Bennington, 1962) (Fig 8). ANOVA with Tukey post hoc tests were conducted to look for significant differences in the timing of blow fly oviposition and life stages. A p-value of 0.05 was used to determine significance.

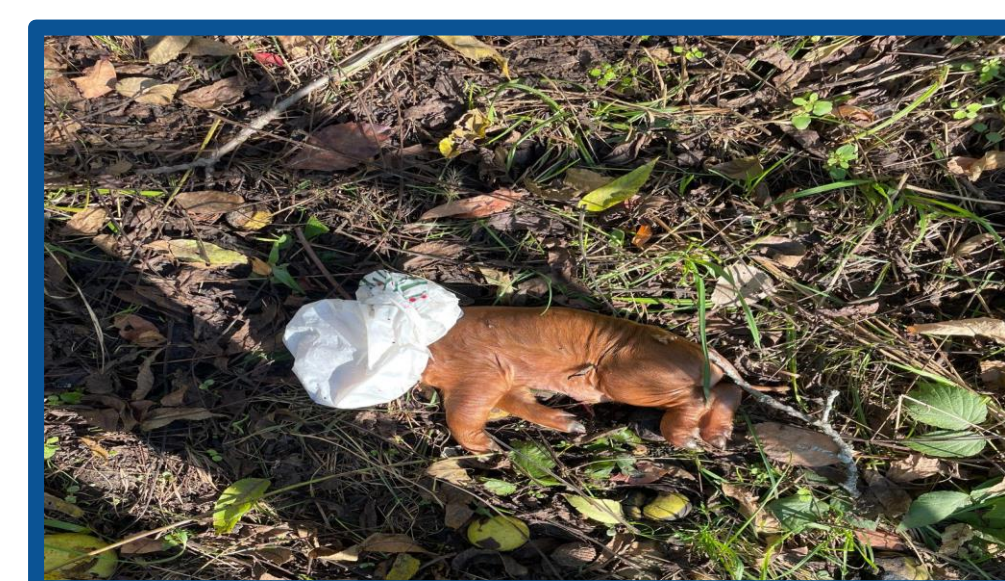


Figure 3. Thick plastic bag covering a pig's head

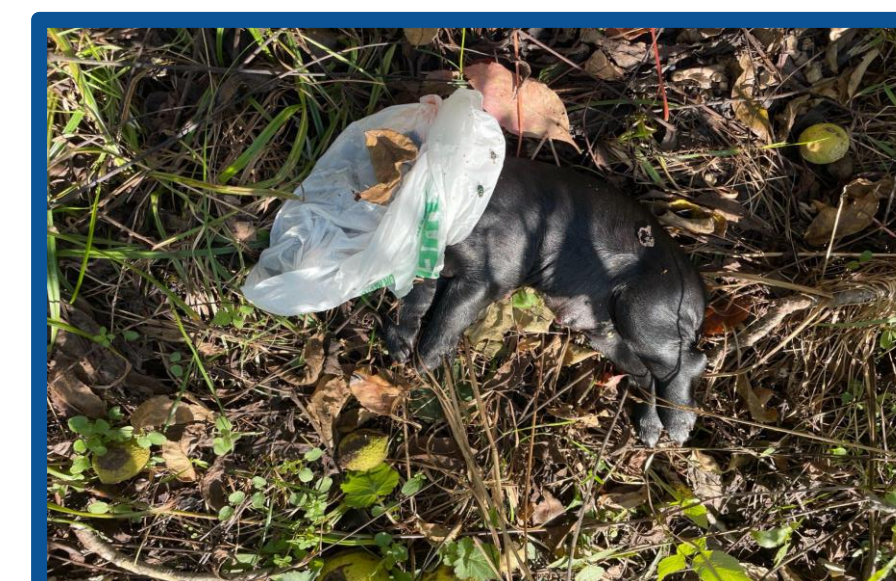


Figure 4. Thin plastic bag covering a pig's head



Figure 5. The field site

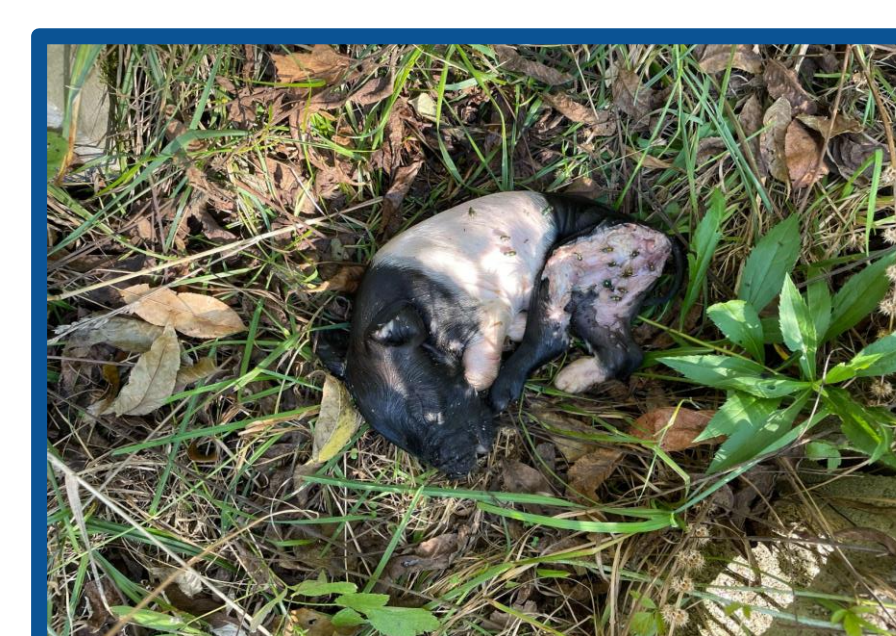
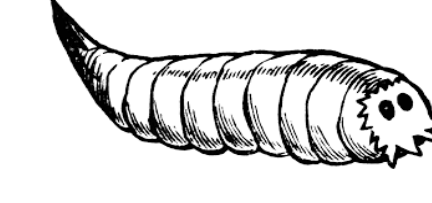


Figure 6. Vertebrate scavenging damage to a pig's hindquarters



Results

- Among all the treatments, *Lucilia coeruleiviridis* (Macquart) was the dominant species found (Fig 7)
- Sarcophagidae *sp.* were not found on controls, but were found on the thin and thick bag treatments (Fig 7)
- *Phormia regina* (Meigen) and *Calliphora vomitoria* (L.) were only found on controls (Fig 7)
- There was no significant difference between the timing of blow fly life events between the treatments
 - The p values for life event timing ranged between 0.22-1

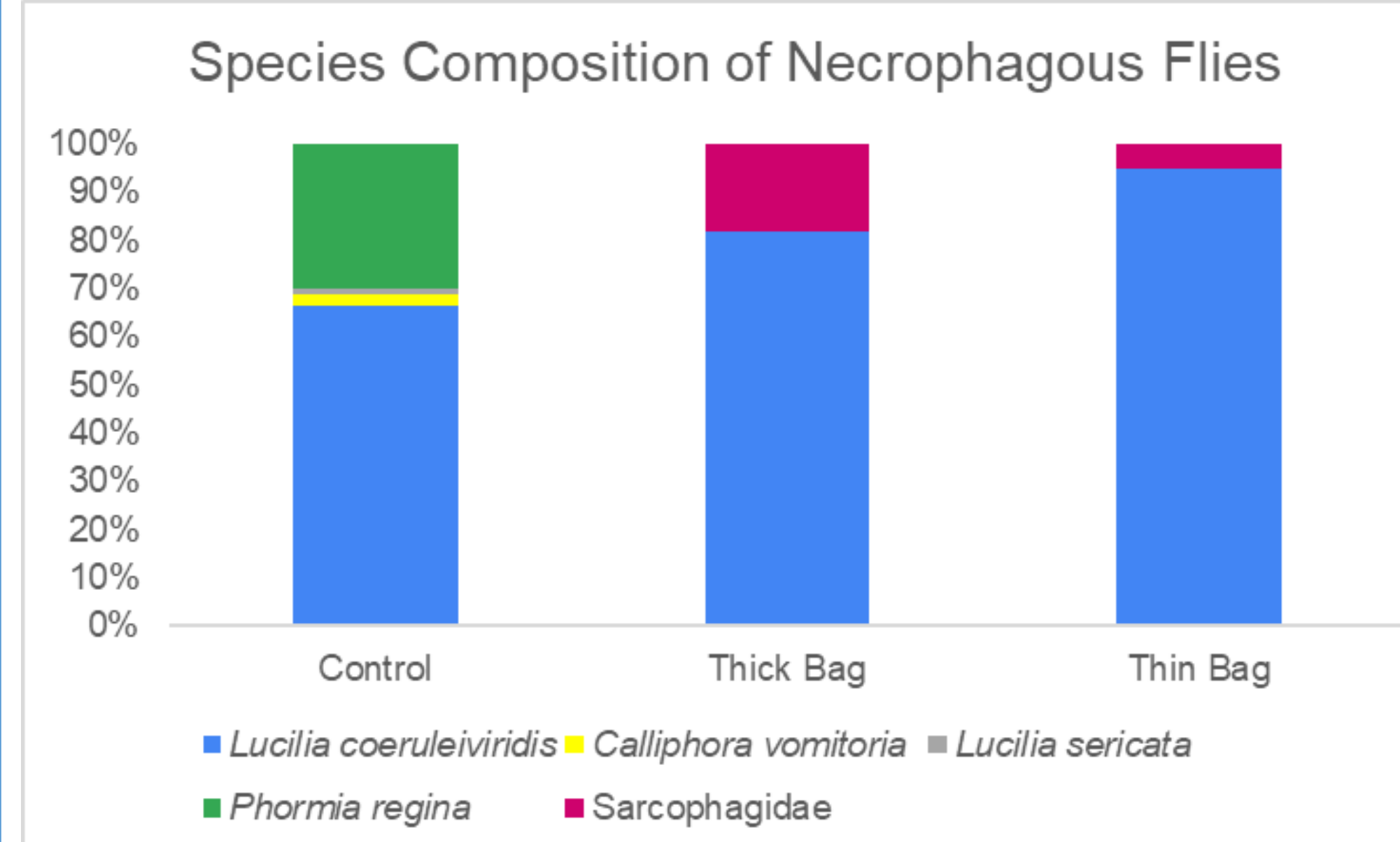
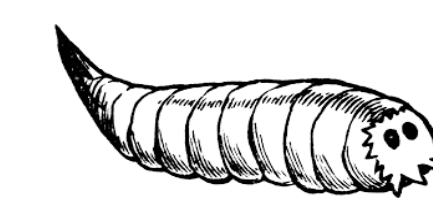


Figure 7 Larval necrophagous fly species composition for control, thick bags and thin bags for two trials

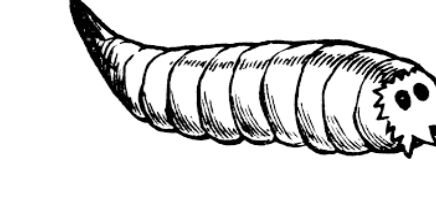


Discussion

The plastic bags did not significantly affect the timing of any of the blow fly life events studied (oviposition, first instar, second instar, third instar, migration). However, the presence of the plastic bag did impact the species composition of necrophagous Diptera, which could impact PMI calculations. Weger *et al.* 2020 studied the effect of a plastic freezer bag on the rate of decomposition and found a decreased rate in head decomposition, but an increased limb decomposition rate. This impacted their PMI calculations. Our results are consistent with this research.

The two dominant flies found (*L. coeruleiviridis* and Sarcophagidae *sp.*) both present problems for PMI calculations. *Lucilia coeruleiviridis* is extremely difficult to rear in a laboratory setting, so the data sets that exist for other species are not available for this fly. It is not possible to identify Sarcophagidae larvae to species because you need to dissect the genitalia of an adult male to gain species specific information. Proper growth tables are essential to accurately estimate the PMI.

Scavenging and rain were problematic in this research, especially in the second trial (Fig 6). Future studies will examine ways to discourage vertebrate scavenging, which impacted the results by displacing pigs/bags during the second trial.



Conclusions

- There were no significant differences found in the timing of Dipteran life events between treatments.
- The species composition of necrophagous flies differed between treatments.
- Sarcophagidae flies were only found on bagged pigs. This was the first time that Sarcophagidae species were collected at this field site in ten years of research.
 - Sarcophagidae larviposit which could explain why the mucous membrane access wasn't as critical.
- Vertebrate scavenging was an issue that needs to be addressed in future studies.

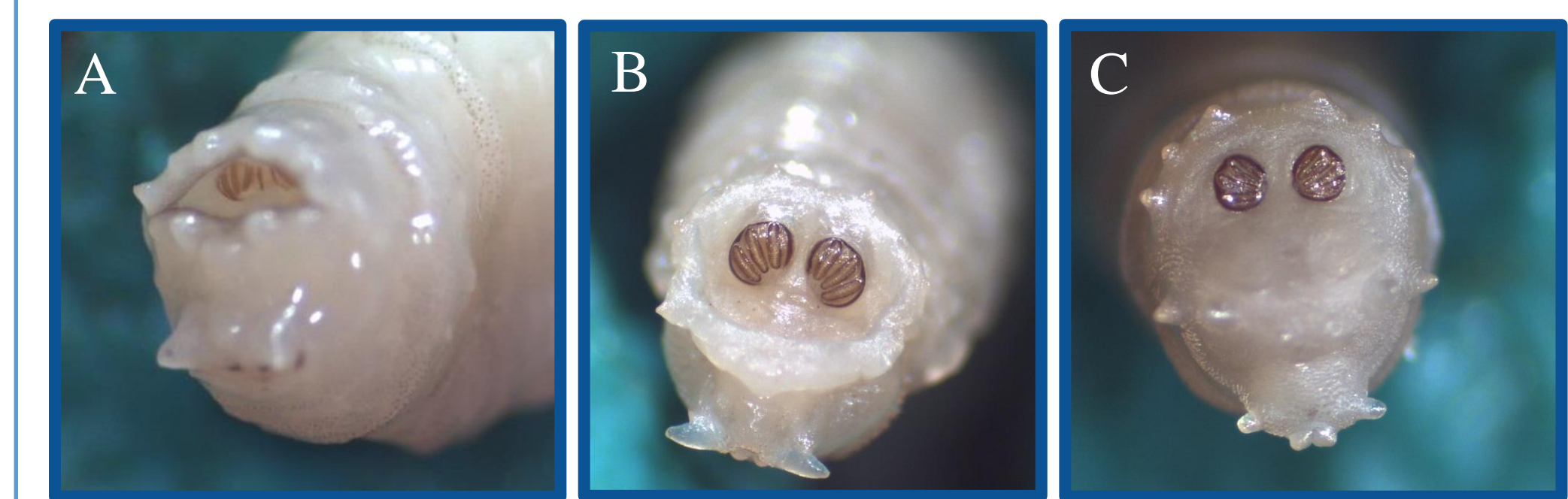
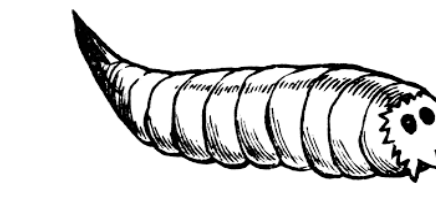
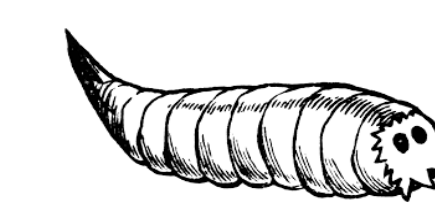


Figure 8. Spiracles viewed through a microscope for three different flies. A. Sarcophagidae, B. *Phormia regina*, C. *Lucilia coeruleiviridis*



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