

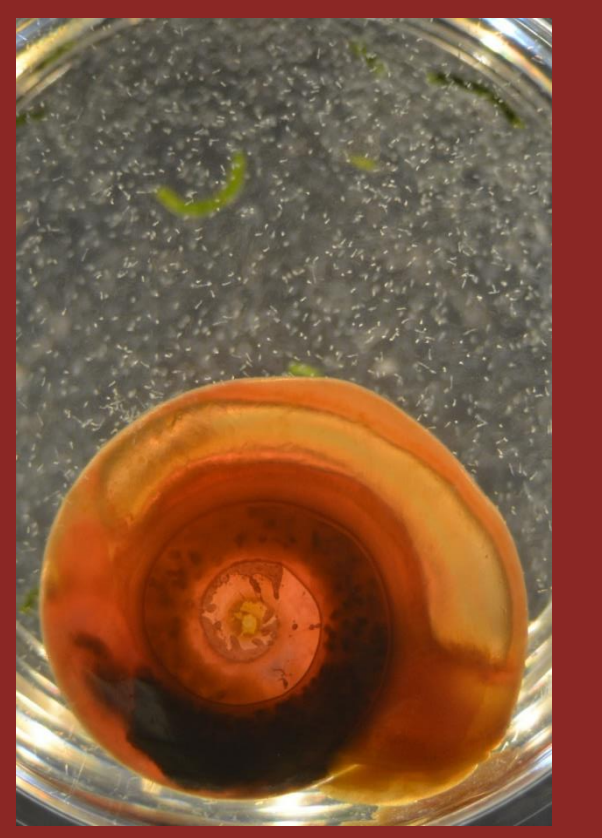


Biomphalaria sudanica

Effect of Diet on the Susceptibility of *Biomphalaria sudanica* to Infection with *Schistosoma mansoni*

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Biomphalaria snail shedding *Schistosoma mansoni*

Objective

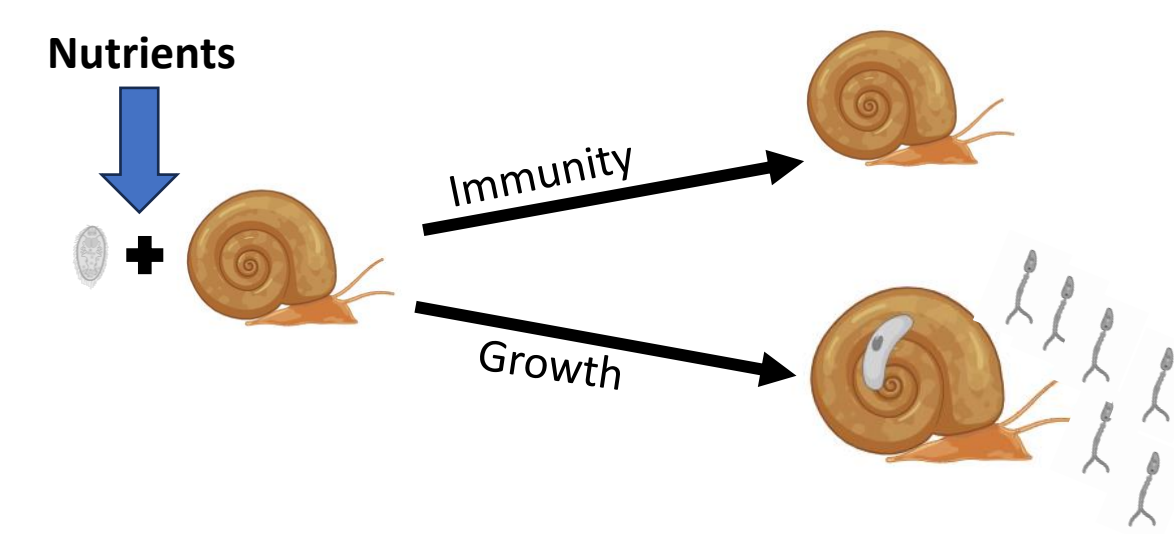
- Determine the role of diet on the susceptibility of African snail vectors to schistosomiasis, an important neglected tropical disease.
- Determine the effect of diet on the amount of cercariae shed by infected snails.

Introduction

Schistosomiasis remains a deadly parasitic disease, with estimates showing 251.4 million people required preventative treatment in 2021 (1). Humans become infected when they contact freshwater containing *Schistosoma mansoni* cercariae that are released from snail vectors. Schistosomes form chronic infections in snails (Figure 1). Once infected, the parasites multiply within the snails and thus a single snail can release hundreds to thousands of cercariae into the environment daily. Therefore, understanding the factors that influence the production of cercariae in a snail is directly related to human infection risk.

Like other snail trematode systems, schistosome infection in snails demands significant energy resources, due to the continuous production of cercariae. Snails also have a resource dependent immune system that is used to fight off a schistosome infection. Thus increased resources could lead to either a boosted immune system capable of withstanding infection, or larger snails with increased cercariae production.

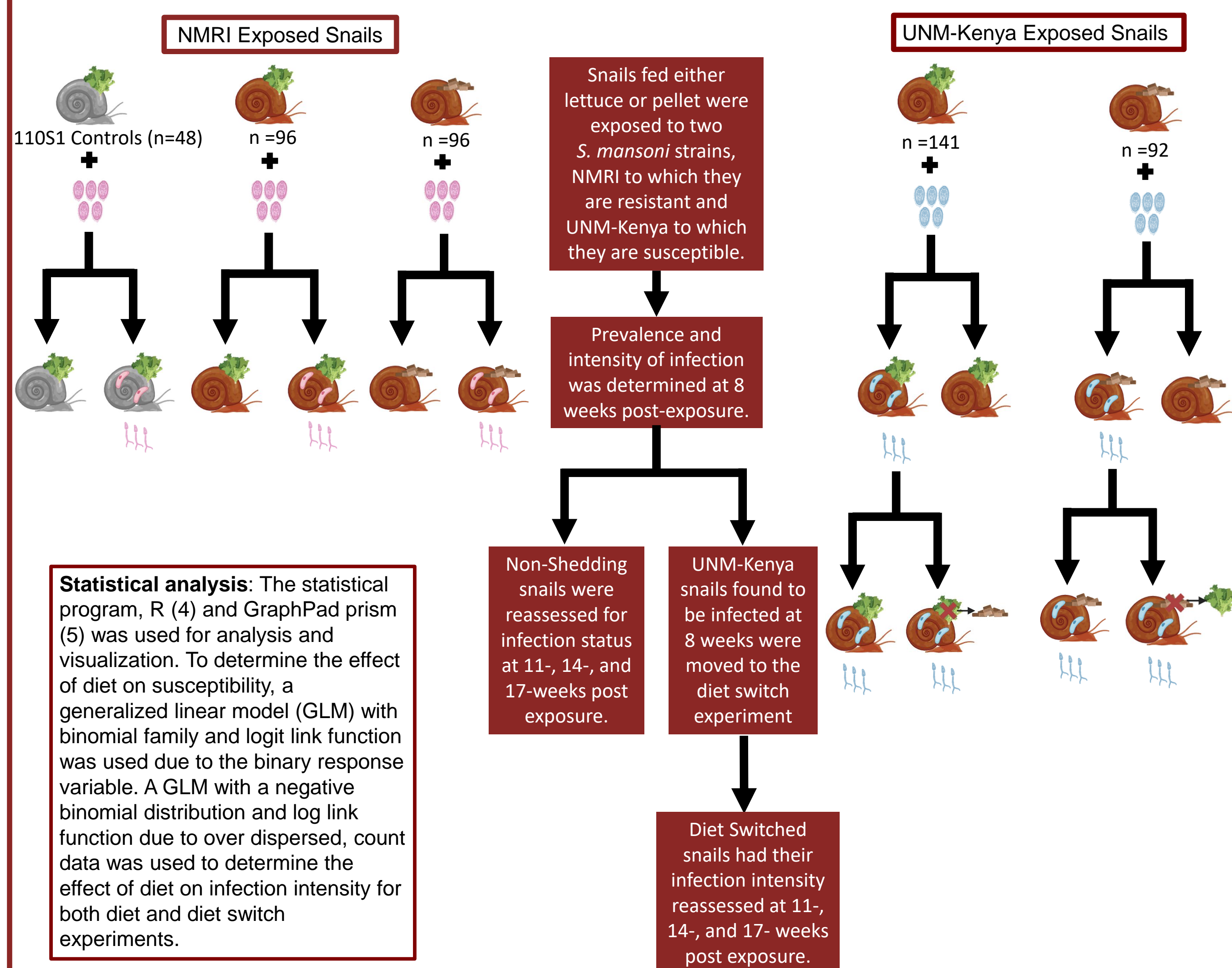
With these potentially opposing forces, the relationship between nutrient availability and parasite transmission is complex.



Methods and Material

Hypothesis

Snails fed the high nutrient diet (pellets)(Figure 2) will be more susceptible to schistosome infection and produce more cercariae than those fed the low nutrient diet (lettuce).



Statistical analysis: The statistical program, R (4) and GraphPad prism (5) was used for analysis and visualization. To determine the effect of diet on susceptibility, a generalized linear model (GLM) with binomial family and logit link function was used due to the binary response variable. A GLM with a negative binomial distribution and log link function due to over dispersed, count data was used to determine the effect of diet on infection intensity for both diet and diet switch experiments.

Results

Does snail diet influence their susceptibility to infection?

Results

Resistant Combination

Snails exposed to NMRI *S. mansoni* showed no significant difference in infection rates between lettuce and pellet diet groups, with 0% and 4% prevalence, respectively (Fisher exact test, $p = 0.2465$). The 110S1 control snails exhibited 44% prevalence ensuring viability of NMRI *S. mansoni* miracidia. (Figure 3)

Susceptible Combination

Snails infected with UNM-Kenya *S. mansoni* exhibited a significant difference between the lettuce and pellet diets with total prevalence of infection for lettuce and pellet fed snails was found to be 32.7% and 47%, respectively (GLM, $P = 0.0304$). (Figure 3)

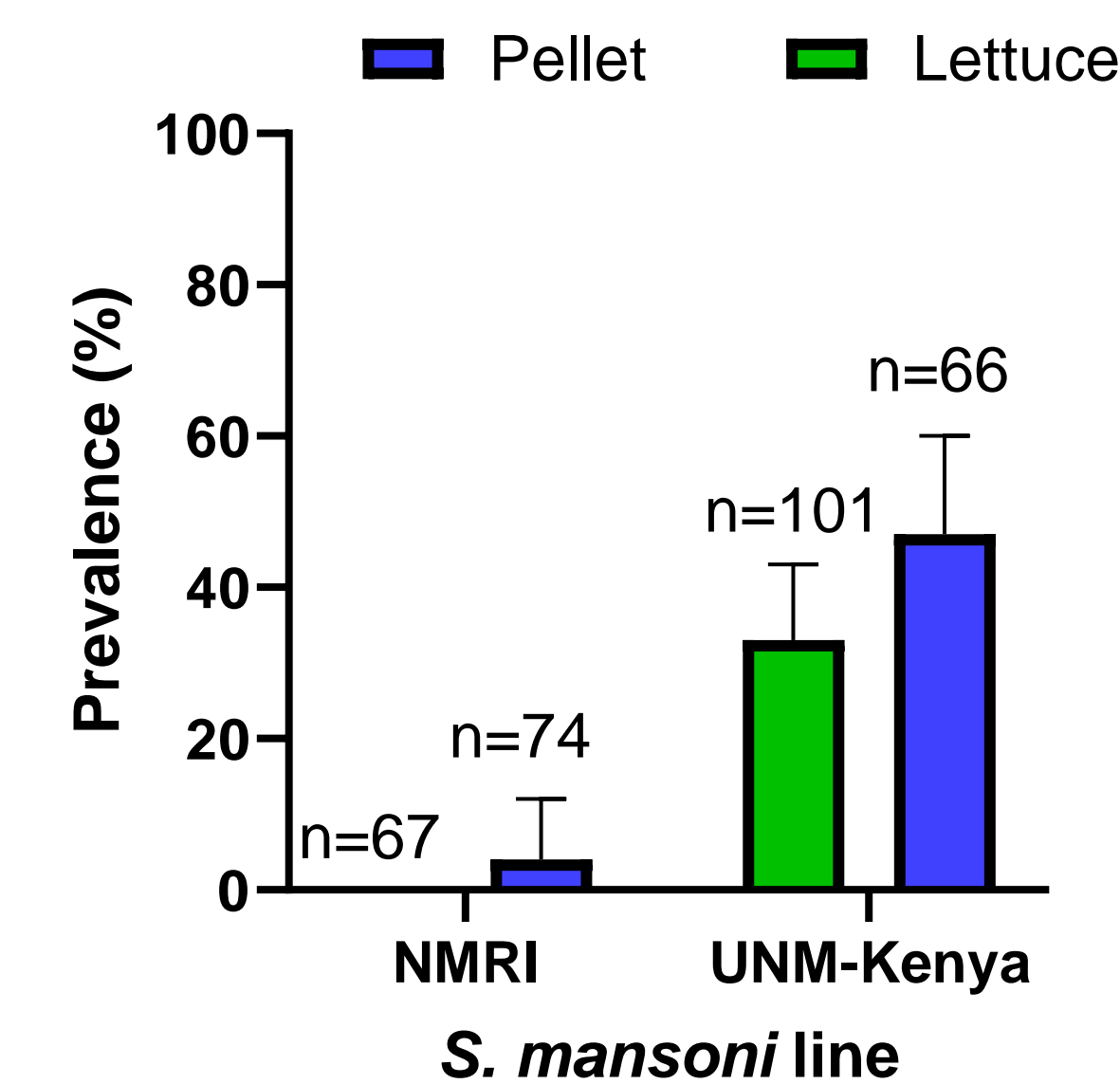


Figure 3: Percent and 95% CI of *B. sudanica* snails fed either lettuce or pellet found to be infected with NMRI or UNM-Kenya.

Does diet impact intensity of infection?

Results

At 8 weeks post exposure snails fed pellets grew larger (LM, $p < 0.0001$) and produced more cercariae than snails fed lettuce at 8 weeks post exposure due to their increased size. (GLM, $P < 0.0001$) (Figure 4) (Figure 5)

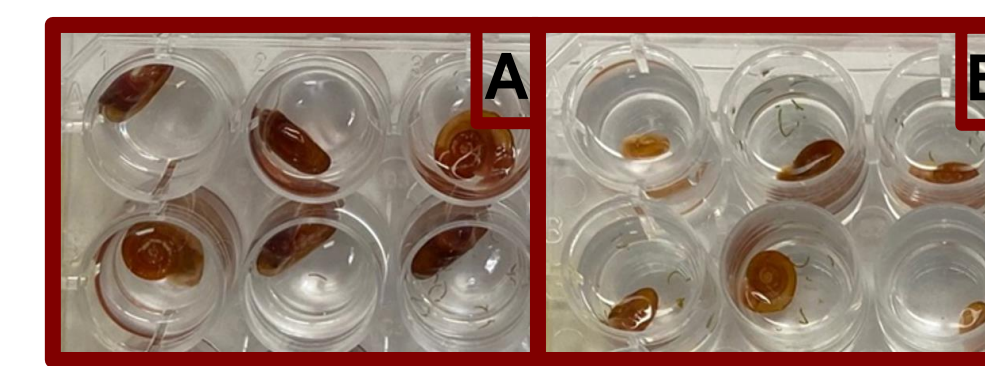


Figure 4: *B. sudanica* snails infected with UNM-Kenya *S. mansoni* fed either pellet (A) or lettuce (B)

Diet Switch

Snails switched from the lettuce diet to the pellet diet had significantly increased cercariae production in comparison to the lettuce control at 11 weeks (GLM, 112%, $P = 0.0285$) and 14 weeks (GLM, 294%, $P = 0.0052$). At 17 weeks there was no significant difference, however sample sizes were small due to mortality of snails.

Interestingly, snails switched from the pellet diet to the lettuce diet initially showed a significant increase in cercariae production in comparison to the pellet control at 11 weeks (GLM, 99% $P = 0.0056$). However, this changed to a significant decrease at 14- and 17- weeks post exposure. (GLM, 14 weeks: 87%, $P = 0.0014$; 17 weeks: 81% $P = 0.0006$).

In contrast to our findings at 8-weeks, there was no significant difference in the number of cercariae shed from snails that remained on a lettuce diet compared to a pellet diet at 11-, 14-, and 17- weeks post exposure (GLM, 11 weeks: $P = 0.5131$; 14 weeks: $P = 0.2907$; 17 weeks: $P = 0.3563$).

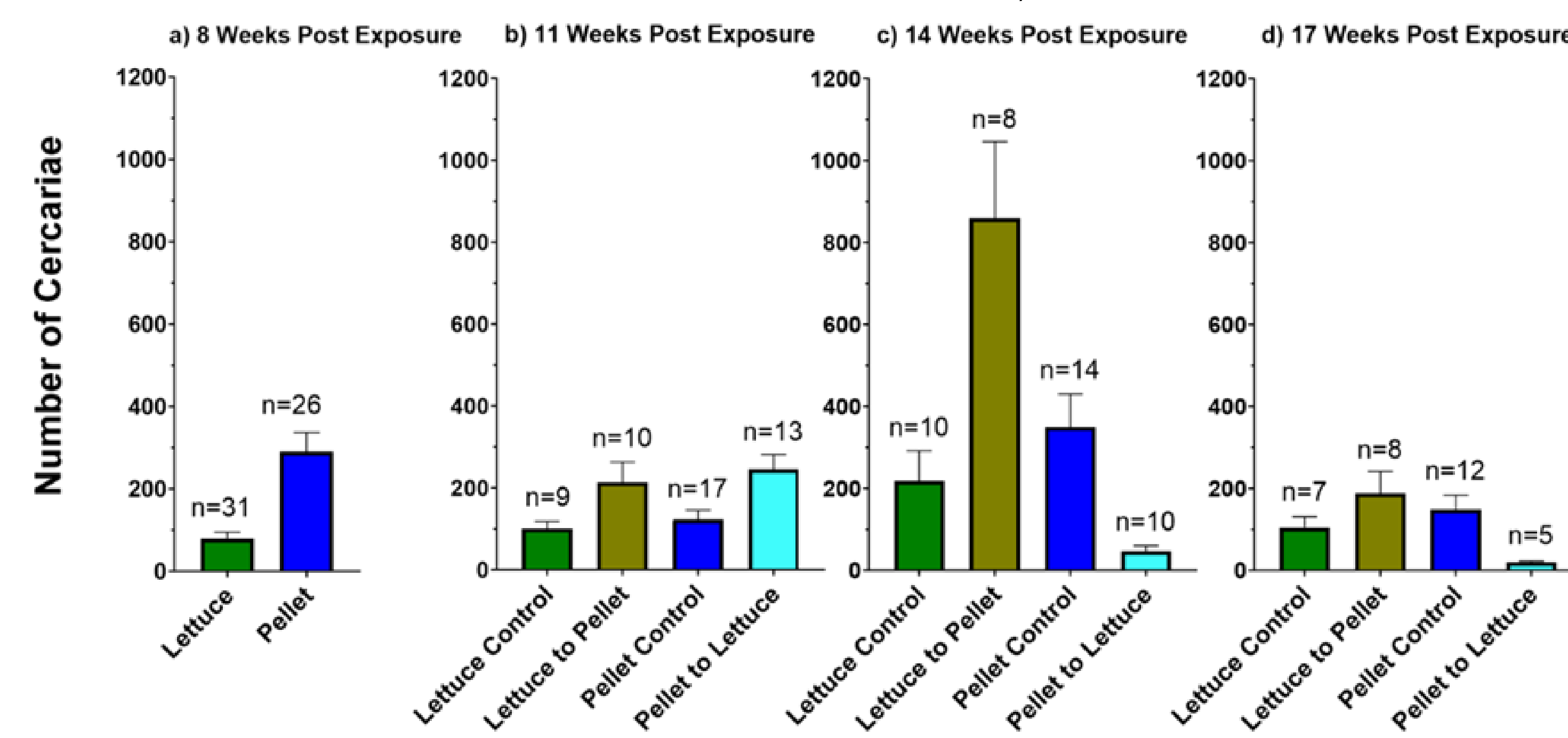


Figure 5: The effect of diet on infection intensity of *Schistosoma mansoni* (UNMKenya line) in *Biomphalaria sudanica* (KEMRIw line). After assessment at 8 weeks, the diet of some snails were switched. Intensity was measured at four assessment intervals a) 8-, b) 11-, c) 14-, and d) 14-weeks post exposure. Error bars represent standard error of the mean and numbers represent the total number of infected snails per group.

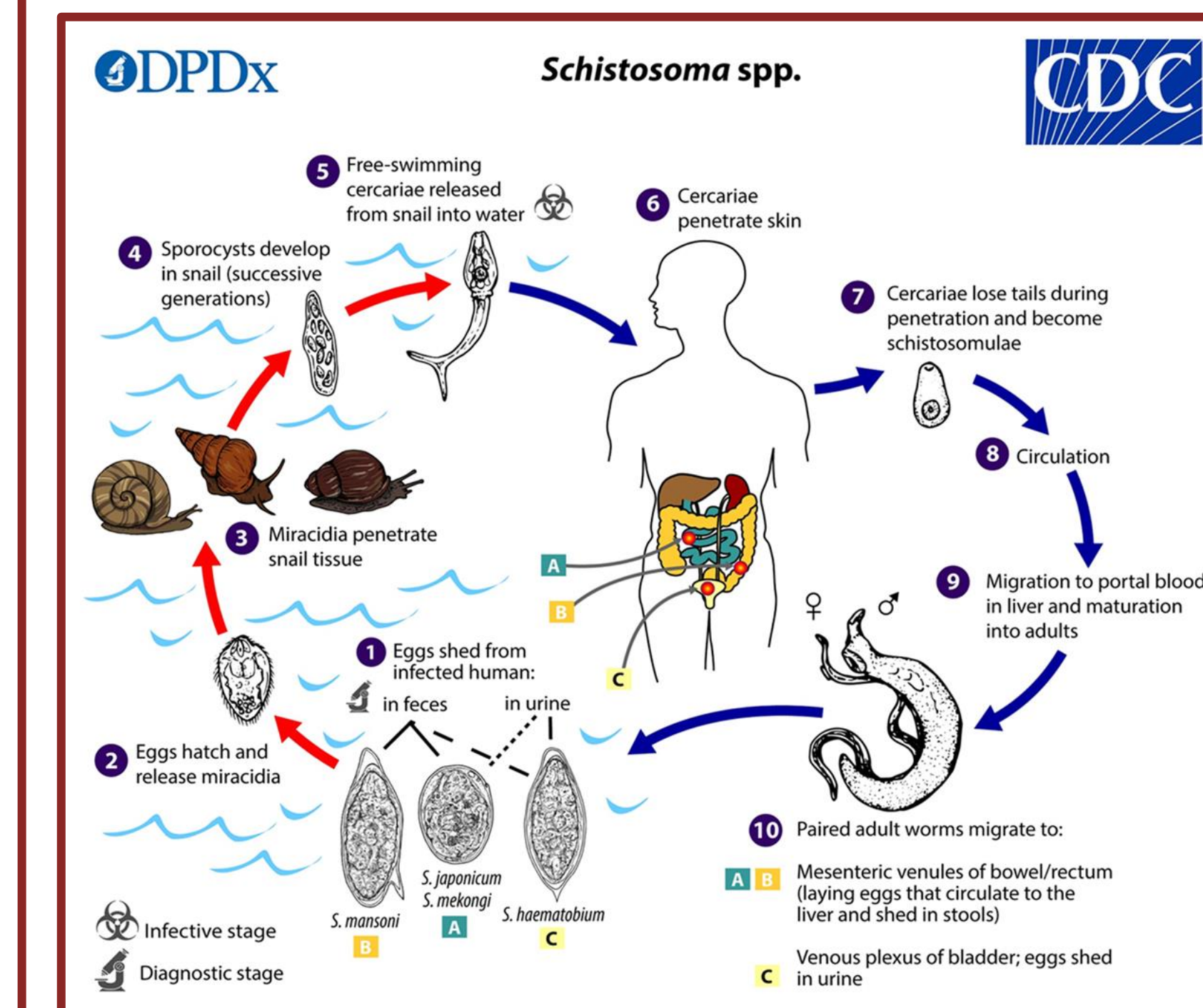


Figure 1: Life cycle of *Schistosoma* spp. (4)



Figure 2: ABF Aquatic Blend Pellet ingredients

Summary

1) **More nutritious diet increased susceptibility of snails to schistosome parasites; however, this did not make resistant snails more susceptible to infection.**

- Pellet fed snails exposed to UNM-Kenya strain of *S. mansoni*, to which they are susceptible, were 101% more likely to get infected than lettuce fed snails.
- Pellet fed snails had higher infection intensity than lettuce fed snails**
- Pellets fed snails produced 295% more cercariae than lettuce fed snails, likely due to their increased size.
- The effect was detected early in infection. But, after 11 weeks, numbers of cercariae were not statistically different.
- Changing infected snails' diet can modulate infection intensity**
- Over time, switching snail diets resulted in an increase in cercariae when the more nutritious pellet diet was given and fewer when the less nutritious lettuce diet was given.
- Interestingly, diet switch resulted in an initial increase in cercarial production even if it was a less nutritious diet.

Conclusion

Our study underlines a complex interaction between *S. mansoni* and its intermediate snail host diet. Results align with our hypothesis that snails fed a more nutritious diet will have higher prevalence of infections and shed more cercariae than their counterparts on the less nutritious diet. Here we demonstrate that snail vitality and its cercariae producing potential can be altered by changes in their nutrition.

This project lays important foundational work towards current snail control efforts that can further our understanding of the epidemiology of schistosomiasis and how environmental and ecological changes may influence human disease risk.

Little is known about snail diets in the wild; however, it is known that macrophytes, diatoms, benthic algae, and detritus are all viable food sources for snails (6) Furthermore, all these species are vulnerable to changes in environmental conditions such as pollution or eutrophication. Therefore, one can conclude that schistosome transmission is a whole ecosystem process, and more work will need to be done to fill in our knowledge gaps.

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