

Differences in abundance of *Microtus pennsylvanicus* between wetlands with
different grazing histories

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Abstract

Grazing by large herbivores can decrease vegetative food resources and reduce cover from predators, causing a decline in small mammal abundance. Conversely, the removal of large herbivores can result in an increase of small mammal density, which might also increase the parasite intensity of these small mammals. Small mammals in an ungrazed area may be more heavily parasitized due to increased contact rates with conspecifics and greater overlap of territory. This study assessed differences in *Microtus pennsylvanicus* densities, as well as the diversity and intensity of their parasites, between lands with different grazing histories on the Flathead Reservation administered by the Confederated Salish and Kootenai tribes of Montana. I hypothesized that there will be greater densities of *Microtus* on ungrazed lands due to either increased cover or plant biomass and that these mammals in ungrazed lands will experience greater parasite diversity and intensity. A trend toward more *Microtus* on more lightly grazed land was indicated though shown to be statistically insignificant and vegetative structure seemed to influence *Microtus* populations in some indeterminate way. Too few parasites were collected to examine the relationship between host density and parasite loads.

Introduction

Effects of grazing on small mammals

Numerous studies have found that small mammal populations increase with the exclusion of large herbivores. (Caro 2002, McCauley et al 2006, Heske and Campbell 1991) Small mammal abundances increase within exclosures which excluded larger herbivores in Botswana (Saetnam and Skarpe 2006) and both abundance and richness increase outside of the Katavi National Park, where the land is less heavily grazed than inside the park. (Caro 2002) The abundance of the pouched mouse (*Saccostomus mearnsi*) is greater in studies in which large herbivores were excluded. (McCauley et al 2006, 2008, Keesing 1998). Similar results were found in Norway, when sheep populations were reduced and field voles (*Microtus agrestis*) became more numerous. (Steen et al 2005) This trend holds true for North American systems as well. Bock et al (1984) noted an increase in rodents in the absence of cattle in semidesert grassland in Arizona. Small mammal abundances were also greater in the Chihuahuan desert in the absence of grazing

(Heske and Campbell 1991) as well as on Arizona wetlands. (Hayward et al 1997) The effects of grazing on small mammals are also seen in bunchgrass, shortgrass and tallgrass habitats across the western United States (Grant et al 1982) and for several species of small mammals in Oregon woodlands. (Johnston and Anthony 2008)

The data is mixed as to how large ungulate grazing limits small mammal populations. One hypothesis is that large herbivores significantly reduce vegetative cover, exposing small mammals to predation. In some instances ungrazed areas were found to have more vegetative cover due to increased leaf litter, standing dead grass (Saetnam and Skarpe 2006), and live grass height (Saetnam and Skarpe 2006, Bock et al 1984). Another theory is that large herbivores compete with small mammals for food, and thus their absence results in relaxed competition. Steen et al (2005) found that while *Microtus agrestis* became more populous when sheep were removed, bank voles (*Clethrionomys glareolus*) did not. This may be because *M. agrestis* shared the diet of the sheep, but *C. glareolus* did not. Additionally, small mammals in ungrazed sites are shown to have a greater body mass, suggesting a better diet. (Keesing et al 1998) Finally, Caro (2002) failed to find any association between either small mammal abundance or richness with vegetative cover (2002). These conflicting results indicate that perhaps both factors—protection from predators and reduced competition for food—affect small mammal densities and that their relative importance varies from habitat to habitat.

Relationship between host density and parasite intensity and diversity

A theory proposed by Altzier et al (2003) states that host density and parasite loads should be positively correlated, due to increased contact with conspecifics leading to a greater opportunity for transmission of parasites. It is generally true that the prevalence and intensity of strongylid parasites increase with host density when considering mammalian species (Arneberg et al 1998) and that gerbils experience a heavier intensity of fleas as their density increased. (Krasnov et al 2002) Furthermore, when animals form social groups, individuals in larger groups frequently face greater exposure to parasites and disease. (Cote and Poulin 1995, Freeland 1979, Moore et al 1988, Davies et al 1991, Hoogland 1979, Brown and Brown 1986)

However, there are instances when host density and parasite intensity become decoupled. As stated above, *S. mearnsi* lives in greater abundance on ungrazed lands, but ectoparasite prevalence and intensity in these areas did not increase as was expected. (McCauley et al 2008) There are a number of potential explanations for this. Ezenwa (2003) saw a relationship between group size and parasite loads only when the groups were stable and the parasite was host specific. Imbalances in host demographics can limit the average parasite intensity in a population, as in the case of fleas parasitizing rats. It was shown that males and larger rats have more parasites than females and smaller ones. (Soliman et al 2001) Environmental factors may also play a role; tapeworm prevalence increased but intensity decreased for sand rats during the dry season. (Fichet-Calvet et al 2002) There may also be competition between different parasite species attenuating parasite abundance on more densely packed hosts. (Day and Benton 1998, Krasnov et al 2005, Stanko et al 2006) Thus, there are then many factors besides host density that come into play to determine parasite intensity and diversity.

Hypotheses

This study examined the small mammal population on the Confederated Salish and Kootenai tribal lands in western Montana, seeking to characterize the effects of grazing on small mammal populations and whether there is greater parasite diversity and intensity with greater host density. I focused my analysis on the meadow vole, *Microtus pennsylvanicus*, the most abundant small animal at my study site this year. *Peromyscus maniculatus* is also numerous in the area, but was not captured in great enough numbers to analyze a response to grazing. I hypothesized that there will be more *Microtus* in less heavily grazed land and that this will correspond with either greater plant cover (equating to diminished risk of predation) or greater plant biomass (signifying relaxed competition for food resources) or both. In the same vein, I hypothesized *Microtus* on less heavily grazed land will have a greater average weight, signifying greater availability of food.

I also hypothesize that there will be greater parasite diversity (i.e., number of parasite species) and intensity (i.e., total number of parasites per individual host) and prevalence (i.e., number of infected

host individuals in the population) in less heavily grazed areas owing to the increase in small mammal density.

Methods

Study site and species

The study took place on nine sites on the Ninepipes National Wildlife Refuge and on the National Bison Range in Charlo, Montana. These areas are all low-lying areas by wetlands with three different grazing histories. Crow, Johnson 80 and one site on the National Bison Range were grazed in the current year, 2011. Another three sites, one at Herak and two at Sandsmark WPAs, were grazed in summer 2010. The final three sites, one at Montgomery WPA and two on tribal trust lands off Leon Road, have not been grazed recently. Table 1 contains UTM coordinates for the trapping grid at each site.

The two small mammal species trapped in abundance at these sites are *Peromyscus maniculatus* and *Microtus pennsylvanicus*. Both species occur in wetlands and grasslands and can be active at night. *Microtus pennsylvanicus* is primarily herbivorous whereas the diet of *Peromyscus maniculatus* is somewhat more diverse, including a large number of seeds, grains, fruits and insects.

Small mammal trapping and data collection

Small mammals were trapped using collapsible Sherman traps, placed in a 5x5 grid at each site with 10 meter spacing. They were baited with a 1:1 mixture of oats and barley to peanut butter and set in the evening between 1800-2100 h and checked in the morning between 600 and 800 h. Three sites were trapped per night, one from each of the three possible grazing histories. Traps at S1, L1 and J were set out from July 1st until July 21st and were opened for trapping on the nights of the 17th and the 18th. Traps at L2, S2, and C were set out from the night of July 21st until the 27th and were opened for trapping on the nights of the 23rd, 24th, and 26th. Traps were set out at H, M, and NBR from the night of July 27th until August 3rd and were opened for trapping on the nights of the 27th through the 31st. The rodents were marked for identification by painting nails with nail polish, owing to the difficulty of ear tagging *Microtus*. Upon capture, the species, sex, weight and length of the animal were assessed and recorded, as well as the number and location of recaptures.

Vegetation assays

Vegetation measurements were taken from four subplots, one per corner in the grid created by the traps, and averaged for each site. Percent cover of grasses, dead grasses, forbs, bare ground and litter was visually estimated at each sampling point using a 0.5 m² Daubenmire frame. Within the Daubenmire frame I also measured height of grasses, dead grasses and forbs, litter depth, and biomass. Plant biomass for the sites was measured to estimate availability of food using a Robel pole.

Ectoparasite analysis

For the ectoparasite analysis, both fleas and ticks were assessed. For fleas, the animals were brushed with a flea comb. To keep methods consistent and results quantitative, each animal was held over a shallow basin with a small amount of ethanol and brushed ten times down the right flank from neck to tail. Only fleas stuck in the comb or dislodged into the ethanol were to be counted. (McCauley et al 2008) and taken back to the lab for identification. All nymphal and larval ticks visible on the body and head were to be counted and identified to species. (Brunner and Ostfeld 2008)

Endoparasite analysis

Feces were collected from the trap each morning, bagged and taken back to the lab. They were assessed using the McMaster technique. As many droppings as could be collected were weighed and mixed with 30 mL of Sheather's sugar solution in a fecal cup. Fifteen mL of the mixture were placed into a centrifuge tube with another 15 mL of sugar solution. The two were mixed by gently rocking the centrifuge tube back and forth. A portion of the fecal-sugar solution mixture was pipetted into the chambers of the McMaster slide. After allowing five minutes for the eggs and oocysts to rise to the top, the slides were examined under a microscope. The columns on the chambers allow the parasite eggs to be quantitatively counted. After all the eggs in both chambers are counted, that number is converted to the number of eggs per gram of feces.

Statistics

All statistical tests were carried out in R 2.31.1. A Friedman Rank Sum Test was performed to compare *Microtus* capture success between the three different grazing schemes. The days they were

trapped were used as the blocks to remove variation from weather and length of trapping period. A Pearson's Correlation Matrix and Principle Components Analysis were run to determine components to explain the variation in vegetation measurements between sites. These principles components were then used as factors in a Backwards Stepwise Regression with *Microtus* capture success to determine if small mammal populations were in anyway related to vegetation characteristics. Finally, the average weight of *Microtus* between sites was compared using a one-way ANOVA and analyzed in a Backwards Stepwise Regression using the vegetation principle components as factors. Normality of the data was assessed by Shapiro-Wilks tests, Quantile-Quantil Plots and Residual Plots.

Results

Small Mammal Trapping Success

Over the course of the summer, 75 *Microtus pennsylvanicus* were captured and marked by nail polish while 6 were found sickly and released or dead in the trap. Twenty *Peromyscus maniculatus* were trapped and marked, and two *Sorex cinereus* were found dead in the traps. Trap success for each site can be found in Table 1.

Effect of grazing history on Small Mammal Trap Success

A Friedman Rank Sum Test found that differences in small mammal trapping success between sites with different grazing history was just short of statistical significance (Friedman $\chi^2=4.667$, $df=2$, p -value=0.097). The mean trapping success for each type of site is visualized in Figure 1.

A Pearson Correlation Matrix was run on 11 vegetation variables to determine if any were highly correlated. Grass height, dead grass height and the Robel Pole Measurements of vegetation density were all highly correlated ($r \geq 0.712$) so only the Robel Pole Measurements were retained. Percent litter cover and percent dead grass cover were also highly correlated ($r=0.94$) so percent dead grass cover was dropped. The remaining variables were entered into the Principle Component Analysis and included percent litter cover, percent grass cover, percent forb cover, percent bare ground cover, forb height, litter depth, Robel pole measurements, and the coefficient of variation (CV) of the Robel pole measurements (as a measure of the heterogeneity of the environment). The Principle Component Analysis generated

eight components (See Table 2). The first four (PC1-PC4) explained approximately 88% of the vegetation variation and were those used in further analyses. Table 3 shows which vegetation variables are strongly loaded into each component.

The relationship between trapping success of *Microtus* and the Principle Components was analyzed via a Backwards Stepwise Regression. This concluded that only PC1 predicted *Microtus* trapping success ($R^2=0.4327$, $df=7$, $p\text{-value}=0.032$, see Figure 2). As PC1 is most strongly positively loaded by heterogeneity of the environment (CV of Robel pole measurements) and percent forb cover and most negatively loaded by percent grass cover, this suggests that *Microtus* populations may also be influenced by heterogeneity, forb cover and grass cover. Normality of the data was confirmed by a Shapiro-Wilks test ($W=0.986$, $p\text{-value}=0.9879$) and by a residual plot and Quantile-Quantile Plot (See Figure 3).

The average weight of *Microtus* at sites of each of the three grazing histories was tested for normality by a Shapiro-Wilks ($W=0.911$, $p\text{-value}=0.323$) and analyzed with a one-way ANOVA. No significant difference between grazing histories was detected ($F=0.317$, $df=2$, $p\text{-value}=0.74$). The average weight data was then tested in a Stepwise Backwards Regression with the Principle Components as factors, but no significant relationship was apparent ($R^2=0.128$, $df=5$, $p\text{-value}=0.347$).

Parasite Tests

Four of the *Microtus* captured had nymphs of the ravaging fever tick *Ornithodoros hermsi* and two had nymphs of the wood tick *Dermacentor andersoni*. These were found during laboratory examination of the debris combed off the mammals in the field. No other ecto- or endoparasites were observed. With so few data, it was not possible for statistical analysis of parasitism trends.

Discussion

In numerous other systems around the world, small mammal populations are higher on ungrazed land than in areas that are grazed by large herbivores. (McCauley et al 2006, Steen et al 2005, Bock et al 1984) In several of these systems, it was theorized that this was due to reduction in vegetative cover by large herbivores, exposing small mammals to predators. (Saetnam and Skarpe 2006) Another possibility

is that there is diminished competition for herbaceous food on ungrazed lands and a number of studies have found evidence supporting this theory. (Caro 2002, Keesing 1998, Steen et al 2005) The current study investigated the effects of different grazing schemes on the meadow vole, *Microtus pennsylvanicus*, on the Ninepipes Wildlife Refuge and National Bison Range in western Montana. *Microtus* are herbivorous rodents that might to be particularly sensitive to effects of grazing, especially in low lying areas (Johnston and Anthony 2008). Additionally, they are prey to a number of carnivores (raptors, weasels, foxes) and so would potentially benefit from increases in vegetative cover. (Reich 1981) It does appear that there are greater numbers of *Microtus* on land that had not been frequently or recently grazed though this trend was not statistically significant. It is possible that a larger sample size or more trapping data would yield a significant result in concurrence with previous studies in Africa (Caro 2002, McCauley et al 2006) and other parts of North America. (Bock et al 1984, Heske and Campbell 1991)

Additionally, the relationship between *Microtus* populations and a number of vegetation measurements, including percent cover, height of grasses and forbs, litter depth and density, was analyzed to see what attributes of the flora small mammal populations might be sensitive to. A principle component analysis of the vegetation variables generated a component that was significantly correlated with *Microtus* numbers. This component was heavily positively loaded with percent cover by forbs and heterogeneity of vegetation density in the environment and negatively loaded by percent cover of grass. It is logical to presume that as percent cover by forbs increase, percent cover by grass would decrease and it is conceivable that greater cover by forbs would increase the heterogeneity of the environment, seeing as forbs are less uniform in their coverage than grass. How this would positively affect *Microtus* populations is unclear, though *Microtus* do prefer areas with greater cover. (Reich 1981) It may also be that *Microtus* prefer moister environments (Reich 1981) and that forbs grow better in moister areas. To casual observation it did seem that some of the more heavily populated sites were on wetter land. While further investigation is needed to tease apart exactly what factors of vegetative structure can influence *Microtus* populations, it appears as if the floral community does have some relationship with small mammal numbers, as previously supposed. (Grant et al 1982)

Keesing (1998) reported that small mammals in ungrazed land had on average higher weight than in grazed, possibly indicating that they were able to feed more in the absence of large grazers. However, I did not observe this trend in my data.

Another possibility is that grazing has effects which alter the dynamics of inter-species competition. Though there was not enough data to analyze statistically, the author noticed that at sites with less *Microtus*—usually sites with more recent grazing—there were more *Peromyscus* trapped, and that no *Peromyscus* were trapped at sites with very high numbers of *Microtus* and more infrequent. Because we were not able to run tests on *Peromyscus* data, and because *Peromyscus* historically have had very varied response to grazing by large herbivores (Bock et al 1984, Heske and Campbell 1991, Clary and Medin 1991, Grant et al 1982, Hayward et al 1997, Johnston and Anthony 2008) it is difficult to speculate exactly how grazing might alter interspecific competition between these two species. However, competition is known to be an important factor influencing *Microtus* populations (Reich 1981) and might be subject to influences by grazing. (Sheen et al 2005)

The parasitological analyses of the captured mammals discovered only a small number of nymphal ticks so the hypotheses relating to how host density is correlated with parasitism rates cannot be answered by this study. It may be that the life cycles of the parasites prevented detection; many gastrointestinal worms are sporadic shedders and many tick species have multiple hosts besides rodents. There is also the possibility that the technique for collecting ectoparasites by combing was not sensitive enough for our purposes and that droppings collected from the traps were not fresh enough for the McMaster's technique to be effective.

It is difficult to say conclusively from this study whether grazing has an effect on *Microtus* populations, though there is reason to believe that this is in fact the case. Similarly, while this study did not precisely illuminate whether grazing altered small mammal populations through changes in the vegetative structure of reduced competition, it did indicate that structure at least probably has some influence on *Microtus* populations. I believe that a greater sample size with more trapping nights will provide the data needed to conclusively answer these hypotheses. This information has important

implications for land management, as voles are important parts of the food chain and their population size has implications for the entire ecosystem. Future studies should also take into account as many species of small mammals in the system as possible, to evaluate the role of competition between species and how grazing might favor one over another. Furthermore, it is recommended that the parasitological studies be revisited at different times of the year and with greater thoroughness to better study the relationship between population size and prevalence and intensity of disease. Both *Microtus* and *Peromyscus* carry parasites that can be transmitted to man, sometimes with severe detriment, so an understanding of the prevalence of disease in small mammal communities will also be useful to public health officials.

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Table 1: Trapping Site Information. Each site, with information on grazing history, UTM coordinates, and trapping success for each species of small mammal. ‘Marked’ mammals are those from who data was collected and marked with nail polish for recapture. ‘Released’ mammals are those who were found sickly but alive and from whom data was not collects.

Site	Grazing Scheme	UTM Coordinates	Trapping Success
Herak	Grazed 2010	0713825 5253377	8 <i>Microtus</i> marked
Sandsmark 1	Grazed 2010	0715795 5254250	18 <i>Microtus</i> marked, 2 released, 1 dead
Leon 1	Grazed Infrequently	0715999 5255004	9 <i>Microtus</i> marked
Leon 2	Grazed Infrequently	0717352 5255442	18 <i>Microtus</i> marked, 1 dead in trap
Sandsmark 2	Grazed 2010	0714202 5254903	10 <i>Microtus</i> marked, 1 released, 12 <i>Peromyscus</i> marked
Montgomery	Grazed Infrequently	0714126 5256345	5 <i>Microtus</i> marked, 1 <i>Peromyscus</i> marked
Crow	Grazed 2011	0710730 5261867	4 <i>Microtus</i> marked, 1 dead, 3 <i>Peromyscus</i> marked, 2 <i>Sorex</i> dead
Johnson 80	Grazed 2011	0711004 5257182	1 <i>Microtus</i> marked, 3 <i>Peromyscus</i> marked
National Bison Range	Grazed 2011	0707997 5249543	2 <i>Microtus</i> marked, 1 <i>Peromyscus</i> marked

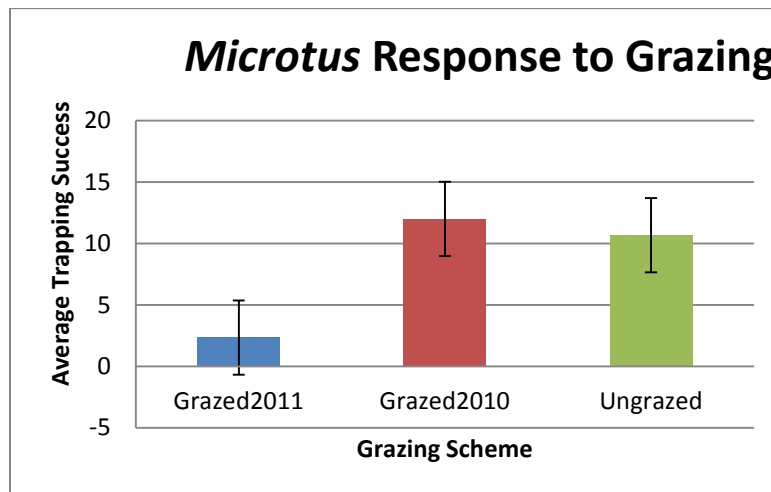


Figure 1: *Microtus* Response to grazing. Mean trapping success for each type of grazing history with standard error. Friedman $\chi^2=4.667, df=2, p\text{-value}=0.097$. Generated in Microsoft Excel.

Table 2: Importance of Principle Components in explaining variation. The relative importance in explaining variation in the vegetation measurements of each principle component are displayed below. The first four explain approximately 88% of the variation and were those used in the following analyses. Generated in R.

Importance of Components:	PC1	PC2	PC3	PC4	PC5	PC6	PC7	PC8
Standard Deviation	1.659788	1.4337	1.1701	0.9257	0.8542	0.3848	0.2646	0.1238
	1	856	247	450	0555	3065	93533	63713
Proportion of Variance	0.344362	0.2569	0.1711	0.1071	0.0912	0.0185	0.0087	0.0019
	1	676	490	255	0839	1183	57833	17777
Cumulative Proportion	0.344362	0.6013	0.7724	0.8796	0.9708	0.9893	0.9980	1.0000
	1	297	787	042	1256	2439	82223	00000

Table 3: Relative loading of vegetation variables into Principle Components. Below are the four components used in regression analyses. Variables that are most strongly loaded into components (either positively or negatively) are in red text on the table. Generated in R.

Vegetation Measurements	PC1	PC2	PC3	PC4
CV Robel Pole	0.4447456	-0.1277262	0.38179525	0.14465067
Forb Height	0.2116880	0.5245664	-0.12130803	0.38219215
Litter Depth	0.3960036	-0.1377325	-0.52409492	-0.39000732
Robel Pole	0.2147826	-0.1797487	0.69615027	-0.04694377
% Bare Ground	-0.2926528	-0.3856719	-0.15362777	0.64504435
% Forb Cover	0.4500180	-0.2156781	-0.20341963	0.48557176
% Grass Cover	-0.4996251	-0.2129494	0.04993946	-0.02103635
% Litter Cover	0.1290428	-0.6454681	-0.11287638	-0.16245380

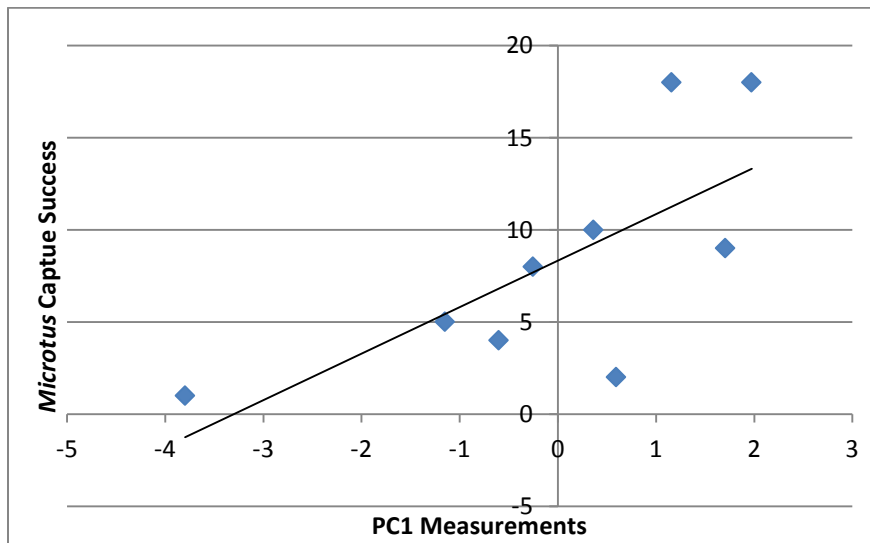


Figure 2: Regression of *Microtus* Trapping Success against PC1. Principle Component 1 positively predicts *Microtus* trapping success according to a Backwards Stepwise Regression ($R^2=0.432$, $df=7$, $p\text{-value}=0.032$). PC1 is most strongly positively loaded by heterogeneity of the vegetation density and percent forb cover and most strongly negatively loaded by percent grass cover. Graph generated in Microsoft Excel.

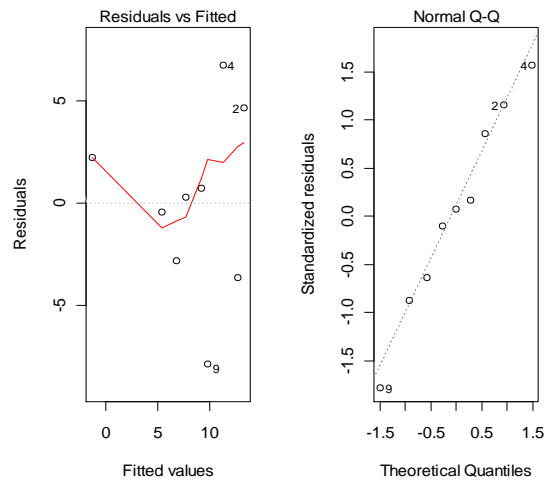


Figure 3: Tests of Normality for the Stepwise Backwards Regression. Plot on the left is a residual plot and plot on the right is a Quantile-Quantile Plot. Data is normally distributed. Graphs generated in R.