Oxalate digestibility in *Neotoma albigula* and *Neotoma mexicana*

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**Summary.** The cactus specialist, *Neotoma albigula*, tolerates high concentrations of potentially harmful oxalate compounds in its diet. Previous research has shown that oxalate compounds are broken down by intestinal microorganisms. Thus the ability of *N. albigula* to utilize a diet high in oxalates may be a consequence of the adaptation of the microflora rather than its own evolution. To test this hypothesis, the oxalate degradation ability of *N. albigula* was compared with that of *N. mexicana*, a generalist herbivore. Apparent oxalate digestibility was not significantly different in the two species, when tested using field-acclimated individuals. Analysis of scats recovered from traps indicated that both species were consuming oxalates in the wild. I conclude that the ability of these herbivores to tolerate oxalates is a natural consequence of the utilization of microbial fermentation to degrade the structural carbohydrates of plants coupled with the high adaptive and evolutionary potential of the microflora.

The whitethroat woodrat, *Neotoma albigula*, is almost universally associated with succulent cactus (*Opuntia* spp.) which it presumably depends upon for water (Vorhies and Taylor 1940). *Opuntia* generally contains high concentrations of oxalates (Shirley and Schmidt-Nielsen 1967, and personal observation), which bind calcium as highly insoluble calcium oxalate. As a consequence, a steady diet of cactus could lead to calcium deficiency (Swartzman et al. 1971). Shirley and Schmidt-Nielsen (1967) showed that oxalates in the gut of *Neotoma albigula* are transformed into compounds which move across the gut wall and are then metabolized. By analogy with ruminants, the transformation presumably is accomplished by the intestinal microflora (Dawson et al. 1980).

Shirley and Schmidt-Nielsen (1967) also showed that the sand rat (*Psammomys obesus*), which has a natural diet high in oxalates, can metabolize approximately half as much ingested calcium oxalate as can *Neotoma albigula*; while the hamster (*Mesocricetus auratus*) metabolizes about 1/6 as much as *Neotoma*; and the white rat metabolizes almost none. They suggest that the ability to digest oxalate is an adaptation to a diet rich in oxalates. However, most domestic ruminants can tolerate diets high in oxalates if the microflora of the rumen is exposed to gradually increasing oxalate concentrations (Hodgkinson 1977). In ruminants, at least, the ability to degrade oxalates appears to be an incidental consequence of maintaining a bacterial fermentation chamber for the digestion of structural carbohydrates. Although the chamber in *Neotoma* is in the hindgut rather than the foregut, it is likely that the fermentation process and the responsible micro-organisms are similar (McBee 1977). It is possible that the ability of *Neotoma albigula* to tolerate large amounts of oxalate is ancillary to its utilization of microbes to digest structural carbohydrates. In general, rodents which consume a coarse diet and ferment cellulose in the cecum may be capable of degrading calcium oxalate. Since oxalates are present in low to moderate concentrations in the leafy portions of many plants (Hodgkinson 1977), the microflora could be expected to retain a residual capacity to deal with oxalates even in rodents which do not normally consume a high oxalate diet. The ability of the herbivore to tolerate the secondary compound may not be the result of an evolutionary adaptation of the herbivore to a particular class of secondary compounds, but rather it may derive from the presence of the microbial fermentation chamber and the high adaptive and evolutionary potential of the microflora which resides in it.

In this study I compare the oxalate degradation capacities of field-acclimated individuals of two species of woodrats: *Neotoma albigula*, the cactus specialist, and *N. mexicana*, which Finley (1958) classifies as a generalist herbivore, a species which "shows a marked dislike for cactus" (ibid, p. 423). For comparative purposes, I have also determined the oxalate degradation capability in *Neotoma albigula* acclimated to an artificial diet low in oxalates.

**Materials and methods**

Specimens of *N. albigula* and *N. mexicana* for field-acclimated trials were captured in August 1981 at 3 localities in the vicinity of Showlow, Navajo Co., Arizona (Lone Pine: 13 km N, 6 km W; Mormon Lake: 6 km N, 4 km E; Porter Mountain: 6 km S, 10 km E, all at approximately 2000 m elevation). Vegetation near Showlow was dominated by *Juniperus osteosperma*, *J. monosperma*, and *Pinus edulis* except at Porter Mountain, a riparian situation where *Quercus gambelii* was common. Cactus (*Opuntia whipplei* and *O. plumbea*) was present as scattered clumps at all of these localities, but represented a very small proportion of the biomass. Specimens of *N. albigula* for laboratory-acclimated trials were captured in November 1981 7 km E, 2 km N of Mesa, Maricopa Co., Arizona, elevation 500 m. The
vegetation there was dominated by *Opuntia spp.*, *Cereidium microphyllum*, and *Prosopos juliflora*.

The extent of oxalate degradation was determined by comparing the amount of oxalate consumed in a formulated high-oxalate diet with the amount appearing in the feces. For the field-acclimated trials, woodrats were placed on a diet of whole wheat bread and alfalfa hay pellets with ad-libitum water for one to three days after capture. This was followed by three days on a diet of oxalate-rich bread and ad-libitum hay pellets. In order to avoid contamination of urine and feces with crumbs of oxalate bread, rats were returned to oxalate free bread for a final 24 h period during which urine and feces were collected. Since the passage time through the gut is approximately 24 h in woodrats (personal observation), the feces passed during this period represents the diet during the last day on the oxalate-rich bread. The apparent digestibility, which is defined as the proportion of a substance consumed which does not appear in the feces (Crampton and Harris 1969), was calculated from the average oxalate input per day during the 3 day ingestion period and the oxalate output during the 24 h fecal sampling period.

For the laboratory-acclimated trials, individuals of *N. a. albignula* were held in captivity for eight months prior to testing. During this time they were fed commercial horse pellets (hay with grain), and for 2 months immediately prior to testing, alfalfa hay pellets. This represented a diet low in, but not devoid of oxalates (less than 0.5% of dry matter), such as might be encountered by a generalist herbivore. The experimental regime was the same as that used with the other woodrats. No specimens of *N. mexicana* were available for the laboratory-acclimated trials.

The bread was prepared according to the method of Shirley and Schmidt-Nielsen (1967), in which a dough was made from whole wheat flour, water, and a small amount of sugar, with dried yeast for leavening. After baking at 180°C, the bread was cut into small pieces and dried to a constant weight at 100°C. For the oxalate rich bread, calcium oxalate (ca. 9%) was sifted into the flour before the water was added.

Feces were collected on 3 mm mesh hardware cloth beneath the 13 mm mesh cage bottoms. Urine was collected on household utility paper backed by wax paper. Specimens were air dried, bagged in plastic and stored at room temperature for later analysis.

Scats voided while newly-captured animals were in live traps were air dried and retained for later dietary and oxalate analysis. Dominant dietary items were determined by microscopic examination of one or more fecal pellets after staining with Hertwig’s solution. Some dens of *N. albignula* were dissected and recently gathered vegetation was collected for oxalate analysis. *N. mexicana* dens could not be dissected because of rock over-burden, but common species of vegetation in the vicinity of known *N. mexicana* dens were sampled for oxalate analysis.

Oxalates were determined by the method of Baker (1952). In this procedure, oxalates are solubilized by boiling in 2N HCl, solids are removed by filtering, and proteins precipitated out with tungstophosphoric acid. Oxalates are precipitated out as calcium oxalate and separated by centrifuging, then redissolved in acid and titrated with potassium permanganate solution. The procedure was modified slightly to accommodate different sample sizes, and the wax paper was not boiled, but was instead extracted for a few minutes at room temperature. Blanks run on the wax paper and the utility paper showed no significant oxalate content.

Voucher specimens of *Neotoma spp.* are on deposit in the Museum of Systematic Biology, University of California, Irvine. Species identifications were made by the author and confirmed by R.E. MacMillen.

**Results**

Analysis of variance on the digestibility coefficients from the combined field and laboratory-acclimated trials indicates a significant treatment effect (P<0.001). Comparison of the means with t tests shows that this is due to differences between the mean of the laboratory acclimated *N. albignula* group and the other two (Table 1). The digestibility coefficient for *N. albignula* acclimated to a low oxalate diet in the laboratory, is significantly (P<0.001) lower than that for either *N. albignula* or *N. mexicana*, acclimated to their natural diets. There is no significant difference in the digestibility coefficients for the latter two. That is, the generalist herbivore, *N. mexicana*, degrades oxalates as well as the cactus specialist, *N. albignula*. The amount of oxalate appearing in the urine was between 2 and 11 mg/24 h (about 0.1 to 1% of that consumed, for all species), indicating no appreciable excretion of dietary oxalate in the urine.

The ability of both species to degrade oxalates suggests that they are maintaining an intestinal microflora with oxalate degrading capabilities by consuming oxalates in nature. Oxalate consumption in the natural diet is confirmed by its presence in scats from live traps, as shown in Table 2. Although scats from *N. albignula* yielded significantly more oxalate than did scats from *N. mexicana* (P<0.01), both contain oxalate in amounts indicative of substantial dietary intake, on average. However, a few individuals of *N. mexicana* produced scats very low in oxalate. Four of 13 *N. mexicana* produced scats of less than 1% oxalate, and two of these produced scats of less than 0.1% oxalate. Of course the oxalate content of the diet can be inferred from these data only very approximately, since the relative digestibilities of the other dietary items are unknown, but it would appear that *N. mexicana* is more variable in its consumption of oxalate than is *N. albignula*.

Microscopic analysis of the trap scats also indicates that woodrats are consuming significant amounts of oxalate, on average. The diet of *N. albignula* is dominated by *Juniperus spp.*, with secondary contributions from *Opuntia spp.* and *Sphaeralcea spp.* (Table 2). *Neotoma mexicana* diets are dominated by *Quercus gambelli* at one location and *Juniperus spp.* at another location. As can be seen in Fig. 1, the oxalate content of *Juniperus monosperma* and *Opuntia plum-

| Table 1. Apparent Digestibility of Calcium Oxalate. The laboratory diet contained approximately 0.5% oxalates, expressed as oxalic acid. All digestibilities were determined with a diet of 9% oxalates |

<table>
<thead>
<tr>
<th>Diet Condition</th>
<th>N</th>
<th>Digestibility</th>
<th>s.e.m.</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>N. albignula</em> lab. acclimated</td>
<td>8</td>
<td>0.49*</td>
<td>0.024</td>
</tr>
<tr>
<td><em>N. albignula</em> field acclimated</td>
<td>10</td>
<td>0.72*</td>
<td>0.023</td>
</tr>
<tr>
<td><em>N. mexicana</em> field acclimated</td>
<td>10</td>
<td>0.68*</td>
<td>0.028</td>
</tr>
</tbody>
</table>

* Means bearing different superscript letters differ significantly (P<0.001)