most of the world's protected areas have been established since the early 1970s (1), satellite imagery has been obtained at periodic intervals since 1972, and aerial photographs of many regions date back years or even decades earlier (16), it is also feasible to assess the effectiveness of many protected areas on the basis of their pre- and post-establishment conditions, using the approach presented here. To better understand the effectiveness of protected areas and develop more feasible policies, it is essential to integrate ecology with human demography, human behavior, and socioeconomics (12, 28).

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19. All images were georeferenced to ground control points collected throughout the reserve, using Global Positioning System (GPS) receivers (Pathfinder Pro XRS) with submeter accuracy. Because the Corona data are black-and-white photos, we did a forest/nonforest classification using photo interpretation (or visual classification) to provide a consistent methodology for each of the three time points (1965, 1974, and 1997). The area of interest within each

Corona photo was scanned into a digital image at 1200 dots per inch, giving a ground resolution of approximately 10 m. The individual Corona images were then combined into a single coverage and were classified on the basis of photo interpretation. To enhance the classification accuracies, we used general vegetation delineations on topographic maps, field observation information, interviews with local residents, and observations of areas that were undisturbed over time. The Landsat MSS and TM data were classified with the same procedure used for the Corona photos. To provide consistency, the visual interpretations of the Landsat data were done using false-color infrared images with similar band combinations (MSS bands 4, 5, and 6 and TM bands 2, 3, and 4), and the TM images (30 m by 30 m) were degraded and resampled to a resolution of 80 m by 80 m. To validate the visual interpretations, we surveyed 250 ground-truth plots (the size of each plot was equal to 60 m by 60 m, or 2 × 2 TM image pixels) in the summers of 1998 and 1999 in the reserve, using GPS units (with 1- to 3-m accuracy after differential corrections), and we used digital methods [supervised and unsupervised classifications (16, 18)] based on all four MSS bands and on comparable TM bands to classify the Landsat MSS and TM imagery. The overall correspondences between the digital and visual classifications ranged from 82 to 87%. Based on standard accuracy assessment methods (16), the accuracy for forest classifications of the 1997 TM data using visual and digital methods was between 80 and 88% when the results were compared with the independent data from the ground-truth plots. The visual and digital methods resulted in consistent total forest areas (the forest areas resulting from the digital methods were only 0.61 to 5.76% different from those resulting from the visual method). These testing results indicate that the visual classifications in our study were of high quality and comparability.
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Delayed Compensation for Missing Keystone Species by Colonization

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Because individual species can play key roles, the loss of species through extinction or their gain through colonization can cause major changes in ecosystems. For almost 20 years after kangaroo rats were experimentally removed from a Chihuahuan desert ecosystem in the United States, other rodent species were unable to compensate and use the available resources. This changed abruptly in 1995, when an alien species of pocket mouse colonized the ecosystem, used most of the available resources, and compensated almost completely for the missing kangaroo rats. These results demonstrate the importance of individual species and of colonization and extinction events in the structure and dynamics of ecosystems.

Single species or functional groups of closely related, ecologically similar species can affect the structure and dynamics of ecosystems in several ways: (i) as "mechanical engineers," they can alter physical structure and flows of energy and materials (1); (ii) as predators, parasites, and pathogens, they can affect the dynamics of prey or host populations (2); (iii) as mutualists, they can supply essential resources or services (3); and (iv) as producers and consumers, they can influence the levels and flows of energetic and material resources (4). Species that have large ramifying effects on ecosystems through direct and indirect pathways are often called "keystones" (2, 5, 6). Studies that combine experimental manipulations with long-term moni-

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Fig. 1. Percent of the energy used by kangaroo rats on control plots that was used by small granivores on kangaroo rat removal plots.
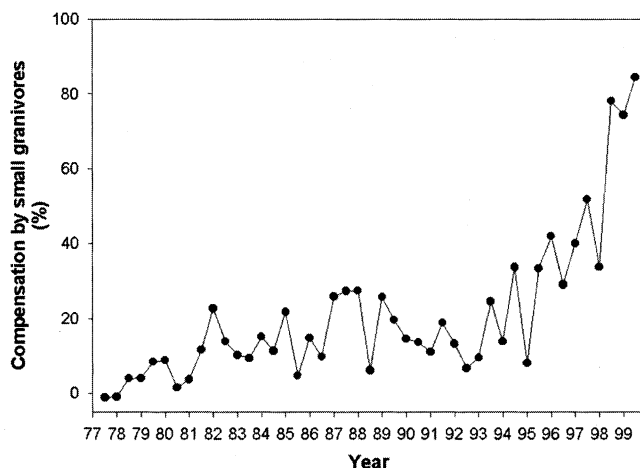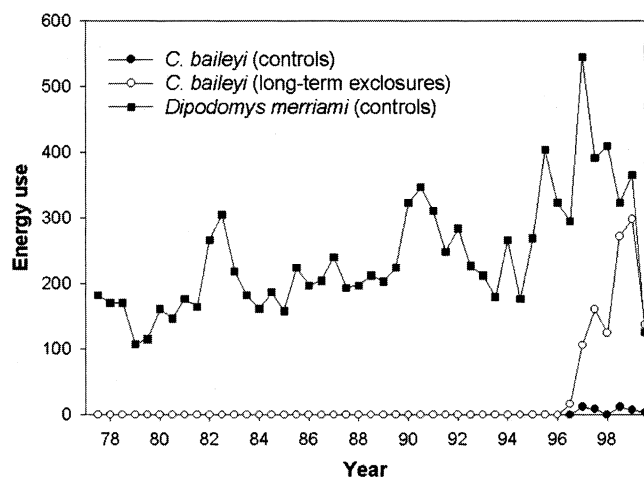

Fig. 2. Energy use (measured in watts) by *C. baileyi* on both the kangaroo rat removal plots and the control plots compared with energy use by *D. merriami*, the dominant kangaroo rat on the control plots for most of the time period.

toring of species populations and their impacts are particularly valuable for assessing the roles of keystones and other species in ecosystems. Studies that document the ecological impacts of removing a keystone species or functional group can answer two important questions: How does the removal of a keystone affect energy flow, species composition, and other aspects of the ecosystem? When these organisms are absent, are other species able to compensate and assume similar roles?

Experiments in the Chihuahuan desert of southeastern Arizona, United States, ongoing since 1977, include the removal of three species of kangaroo rats: seed-eating rodents in the genus *Dipodomys*. Because kangaroo rats are the largest of the seed-eating desert rodents and have enlarged skulls due to their inflated tympanic bullae, it is possible to selectively exclude kangaroo rats while allowing other rodents free access by cutting holes of a calibrated size in fences surrounding experimental plots (7). Control plots are identical in every way, except that they have slightly larger holes that allow all rodents free access, including kangaroo rats. These experiments

have documented two important ecological impacts of kangaroo rats: (i) as dominant seed consumers, they compete with smaller granivorous rodents, suppressing their population densities and altering their foraging behavior and microhabitat use (8–10); and (ii) as selective foragers and agents of disturbance, kangaroo rats have dramatic effects on the composition of annual and perennial vegetation (11, 12). So, by most definitions, kangaroo rats are keystone species in this desert ecosystem.

Until recently, other rodents have been unable to consume more than a small fraction of the food resources that were made available by removing kangaroo rats (8, 13). For nearly 20 years, from the initiation of the experiments in 1977 until 1996, energy use by all of the rodents on kangaroo rat removal plots was consistently very low (Fig. 1), averaging 14% and never exceeding 33% of the energy use by kangaroo rats on the control plots. During this period, eight rodent species have colonized the study site at least temporarily, and six of these species are predominantly granivorous (8, 14). These small seed-eating rodents maintained densities on kangaroo rat

removal plots that averaged almost two times higher than on control plots. Nevertheless, their collective seed consumption failed to compensate for that of the missing kangaroo rats (Table 1).

The situation changed in 1996. A species of pocket mouse, *Chaetodipus baileyi*, never previously observed on the study site, colonized the site and increased rapidly (Fig. 2). *Chaetodipus baileyi* differentially colonized and increased on the kangaroo rat removal plots, where its density averaged nearly 20 times higher than on the control plots where kangaroo rats were present (Table 1). Compensation for missing kangaroo rats, measured as the fraction of the energy used by kangaroo rats on control plots that was used by other rodents on kangaroo rat removal plots, increased from about 33% before 1996 to more than 80% after 1999 (Fig. 1). Since 1998, *C. baileyi* on average accounted for 66% of the energy used by kangaroo rats.

Chaetodipus baileyi is more similar to kangaroo rats in body size and in other aspects of ecology than are the other granivorous rodents that occurred on the study site. Figure 3 compares the distribution of body weights of *C. baileyi*, the other granivorous rodents, and *Dipodomys merriami*, the dominant kangaroo rat that accounted for 66% of the *Dipodomys* and 46% of the granivorous rodents captured on the control plots over the entire time period. Body size correlates with many traits, such as physiology, life history, predation risk, and behavior (15–17). Similarity in body size and correlated traits may explain why *C. baileyi* compensated for the removal of *Dipodomys*. Despite its similarity to kangaroo rats in body size, *C. baileyi* has a substantially smaller skull that allows it to pass through the holes in the fences and colonize kangaroo rat removal plots. The very low density of *C. baileyi* on control plots (Fig. 2) indicates that this species competes strongly with and is nearly excluded by kangaroo rats. This is consistent with other studies showing that *C. baileyi* and *D. merriami* have similar but complementary ecological requirements, so they often replace each other as the dominant seed-eating rodents in nearby habitats that have somewhat different vegetation and soil (14, 18–20).

The long time lag between the removal of the dominant seed-consuming kangaroo rats and the colonization and compensation by *C. baileyi* might be a function of the distance between areas inhabited by *C. baileyi* and our study site. In 1992, the nearest known population of *C. baileyi* was about 5 km away. One individual of *C. baileyi* was captured 1 km from the study site in 1994 (21), the first individual ap-

Table 1. Responses of the six most abundant seed-eating rodents to the removal of *Dipodomys*. Using the 45 6-month periods, energy use on kangaroo rat removal plots was plotted as a function of energy use on control plots. Linear regressions were fitted, slopes were tested for differences from the null expectation of unity, and significantly higher values provide a measure of response to competition.

Species	Slope
C. penicillatus	1.55*
Perognathus flavus	1.28
Peromyscus eremicus	0.90
Peromyscus maniculatus	2.09**
Reithrodontomys megalotis	2.41**
All species except C. baileyi	1.85**
C. baileyi	19.9***

*$P < 0.05$; **$P < 0.01$; ***$P < 0.001$.

Fig. 3. Body size distribution of all individuals of *D. merriami*, *C. baileyi*, and all other non-*Dipodomys* granivores captured at the site.

peared on the site in 1995, and the first evidence of reproduction was in 1996. Regional changes in climate and vegetation during the 1980s and 1990s may have facilitated the colonization (*22*). Since 1997, *C. baileyi* has been the first or second (after *D. merriami*) most abundant rodent species on the site.

Kangaroo rats are keystone species at our study site (*11*). Our long-term study site shows that other species, not recognized as keystones, may be able to assume similar ecological roles. It is important to recognize that compensating species are similar but not identical, and that their ecological roles are complementary but not redundant. *Chaetodipus baileyi* is sufficiently similar to kangaroo rats that it was able to compensate almost completely when kangaroo rats were experimentally removed from our study site. However, kangaroo rats also had large effects on the composition of the plant and rodent communities (*8–12*). It is too soon to know what effects *C. baileyi* will have.

None of the species coexisting within a local ecological community may be able to compensate for a missing keystone. Indeed, none of the 11 species of small seed-eating rodents that were present on our study site before 1996 was able to compensate for the missing kangaroo rats. Compensation occurred only after a time lag of nearly 20 years, when *C. baileyi* colonized the site from a distance of several kilometers. This underscores the importance of long-distance immigration of nonresident species in the response of biota to environmental change (*23*).

We have measured the compensation of *C. baileyi* for missing kangaroo rats in terms of rodent energy use, but the magnitude of compensation must also affect other seed consumers and other trophic levels. Although rodents are the quantitatively dominant seed consumers on our control plots and in most North American desert ecosystems, there are other important granivores (*24, 25*). On the kangaroo rat remov-
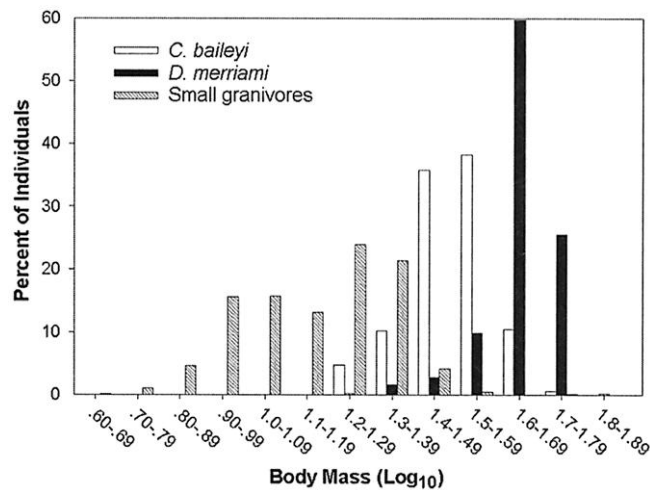
al plots before colonization by *C. baileyi* in 1996, rodents did not consume most of the energy. Energy must have flowed through other organisms, such as birds, ants, or microbes, because seeds did not accumulate. Kangaroo rats also affect the flow of energy through the plant community by their effect on species composition and vegetation structure.

On kangaroo rat removal plots, energy flows to rodent predators probably were also affected as less energy flowed through the rodent community. In summary, the removal of a keystone guild from this ecosystem caused large changes in energy flow and species composition in the rodent and plant communities. The colonization of *C. baileyi* has restored the flow of energy through the rodent community.

Our findings have broader implications, especially regarding the effects of human-caused extinctions of native species and invasions of exotic species. Extinctions of keystone species are likely, almost by definition, to cause major changes in species composition and ecosystem processes. Often the surviving local species will be unable to avert these changes, because they cannot play complementary and compensatory roles. At least partial compensation for missing keystones may sometimes be supplied by species that colonize from distant sources. Any colonists that subsequently become dominant are likely to have their own unique impact on ecological processes, regardless of whether they disperse by their own means or with human assistance, whether they are viewed as "natives" or "exotics," and whether they compensate for missing keystones or have other impacts (*3, 26, 27*). Other recent studies have examined the effects of overall species richness on ecosystem processes (*28–32*). Here we emphasize that the loss of keystone species can drastically alter the species composition and energy flow, and that colonization by a compensating species from a distant source may be necessary to restore ecosystem function. It is important to understand how such colonization and extinction events affect ecosystems if our

response to human-caused environmental change is to be effective and scientifically sound.

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$$\% \text{ compensation} = \frac{E_{sge} - E_{sgc}}{E_D} \times 100$$

where E_{sge} is the energy use of small granivores on the kangaroo rat exclosure, E_{sgc} is the energy use of small granivores on the controls, and E_D is the energy used by *Dipodomys* species on the controls.
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Extension of Life-Span by Loss of CHICO, a *Drosophila* Insulin Receptor Substrate Protein

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The *Drosophila melanogaster* gene *chico* encodes an insulin receptor substrate that functions in an insulin/insulin-like growth factor (IGF) signaling pathway. In the nematode *Caenorhabditis elegans*, insulin/IGF signaling regulates adult longevity. We found that mutation of *chico* extends fruit fly median life-span by up to 48% in homozygotes and 36% in heterozygotes. Extension of life-span was not a result of impaired oogenesis in *chico* females, nor was it consistently correlated with increased stress resistance. The dwarf phenotype of *chico* homozygotes was also unnecessary for extension of life-span. The role of insulin/IGF signaling in regulating animal aging is therefore evolutionarily conserved.

Mutations that extend life-span illuminate the molecular mechanisms underlying aging and longevity. In *Caenorhabditis elegans*, mutation of the genes *daf-2* and *age-1*, which encode components of an insulin/IGF signaling (IIS) pathway, enhances stress resistance and increases adult life-span by up to 200% (*1*). This pathway also controls the formation of dauer larvae, which are developmentally arrested, stress resistant, long-lived, and produced in response to crowding and reduced food (*2*). Potentially, insulin/IGF mutants could be long-lived by virtue of expression of dauer longevity in the adult, in which case the extension of adult life-span by these mutations could be a peculiarity of *C. elegans*. We examined whether the role of IIS in aging has been evolutionarily conserved and therefore might also operate in humans.

In the fruit fly *Drosophila melanogaster*, the insulin/IGF receptor INR, the insulin re-

ceptor substrate CHICO, the phosphatidylinositol 3-kinase (PI3K) Dp110/p60, and the PI3K target protein kinase B (PKB, also known as DAkt1) form a signaling pathway that regulates growth and size (*3–7*). We examined the effects on aging of hypomorphic mutations in *Inr* (equivalent to *daf-2*) and *PKB*, and null mutations in *chico* and the catalytic (*Dp110*, equivalent to *age-1*) and adapter (*p60*) PI3K subunits (*8*). All mutants were tested as heterozygotes. We also tested *chico¹* (*3*) and *PKB³* (*9*) homozygotes and *Inr^{GC25}/Inr^{E19}* transheterozygotes, which form viable dwarf adults. The remaining mutations were homozygous lethal.

Most mutants tested had normal or significantly decreased life-span (*10*). For example, *PKB³* homozygotes and *Inr^{GC25}/Inr^{E19}* flies were short-lived. By contrast, *chico¹* extended life-span (Fig. 1). Homozygous *chico¹* females exhibited an increase of median and maximum life-span of up to 48 and 41%, respectively. *chico¹* heterozygotes also exhibited increases in median life-span of up to 36 and 13% in females and males, respectively. Homozygous males, however, were slightly short-lived.

To confirm that *chico¹* itself extended life-span, we tested the effect on life-span of pCSR4-*chico*, a P element containing *chico(+)*. This construct fully rescues the dwarf phenotype of *chico¹* (*3*). *chico¹* was crossed to two stocks containing independent pCSR4-*chico* in-

sertions (pCSR4-*chico* 1.1 and 2.3). As a control, *chico¹* was also crossed to the base stock in which the P element insertions were made. Progeny with either two copies (*chico¹* heterozygotes with one *chico* transgene) or one copy (*chico¹* heterozygotes alone) of *chico(+)* were compared (*11*). The rescue construct significantly reduced life-span relative to the +/*chico¹* control. The median female life-span of 54 days in +/*chico¹* was reduced to 46 days in +/*chico¹*, +/pCSR4-*chico* 1.1 flies and 52 days in +/*chico¹*, +/pCSR4-*chico* 2.3 flies (*P* = 0.0002 and 0.0243, respectively). Similar effects were observed in males (*10*). Thus, mutation of *chico* itself increases life-span. Because *chico¹* is a null allele, its effect on life-span indicates that the wild-type *chico* gene acts to accelerate aging.

Of the mutations tested, only *chico¹* increased life-span. This may be because the effect of reduced IIS on life-span depends on the degree to which signaling is reduced. Unlike the other null mutations in IIS genes tested, *chico¹* is not homozygous lethal, presumably because the INR receptor can signal to PI3K directly, as well as indirectly via CHICO (*3*). Thus, *chico¹* mutants may be long-lived because of the relatively mild reduction in pathway activity that they bring about. Notably, severe IIS mutations in *C. elegans* can cause premature mortality in some adults, although the maximum life-span of populations is invariably increased (*1*). This is probably why *Inr^{GC25}/Inr^{E19}* flies are short-lived: Demographic analysis indicates that a reduction in the age-specific mortality rate acceleration occurs, whose effect on survival is masked by an elevated rate of age-independent mortality (*12*). Furthermore, a different heteroallelic *Drosophila Inr* mutant to that tested here exhibits an 85% increase in female life-span (*13*). By contrast, in short-lived *PKB³* populations, no reduction in mortality rate acceleration is seen (*12*). This raises the possibility that a second pathway downstream of *chico* might regulate aging in *Drosophila*. Interestingly, CHICO contains potential binding sites for the Drk/Grb2 docking protein, consistent with signaling via Ras/mitogen-activated protein kinase.

We next investigated whether extension of life-span by *chico¹* was mediated by processes previously shown to affect aging. A reduction in fecundity extends life-span in *Drosophila* females (*14, 15*); *chico¹* heterozygous females have reduced fecundity, and the homozygotes are almost sterile (*3, 12*). To test whether the

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