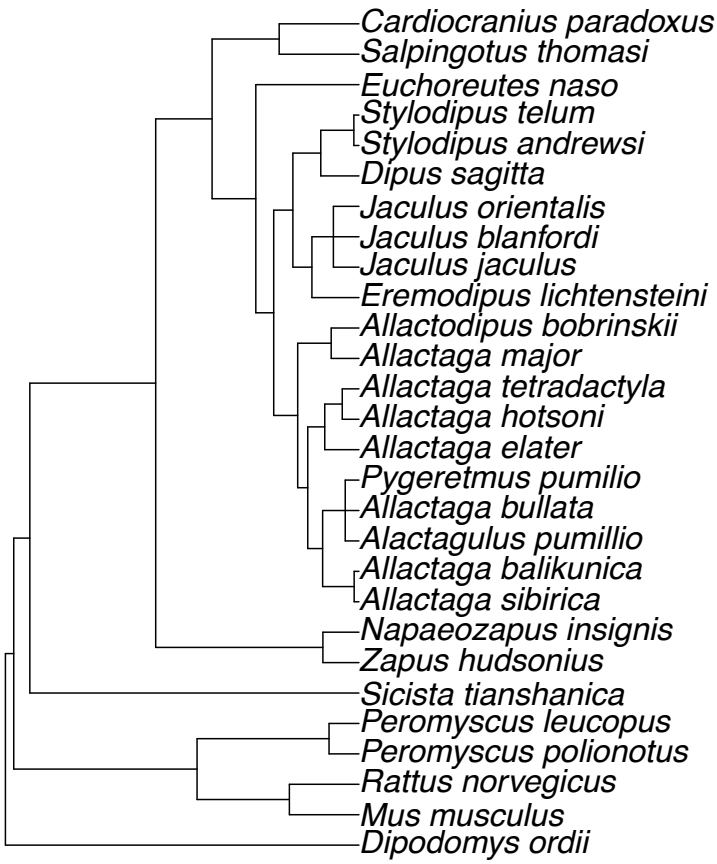

Figure S1: Time-calibrated phylogeny of Dipodoidea generated by processing the Nexus file below using Mr. Bayes. 23 Dipodoid and 5 outgroup taxa were included in this phylogeny. Supports Figure 3B.

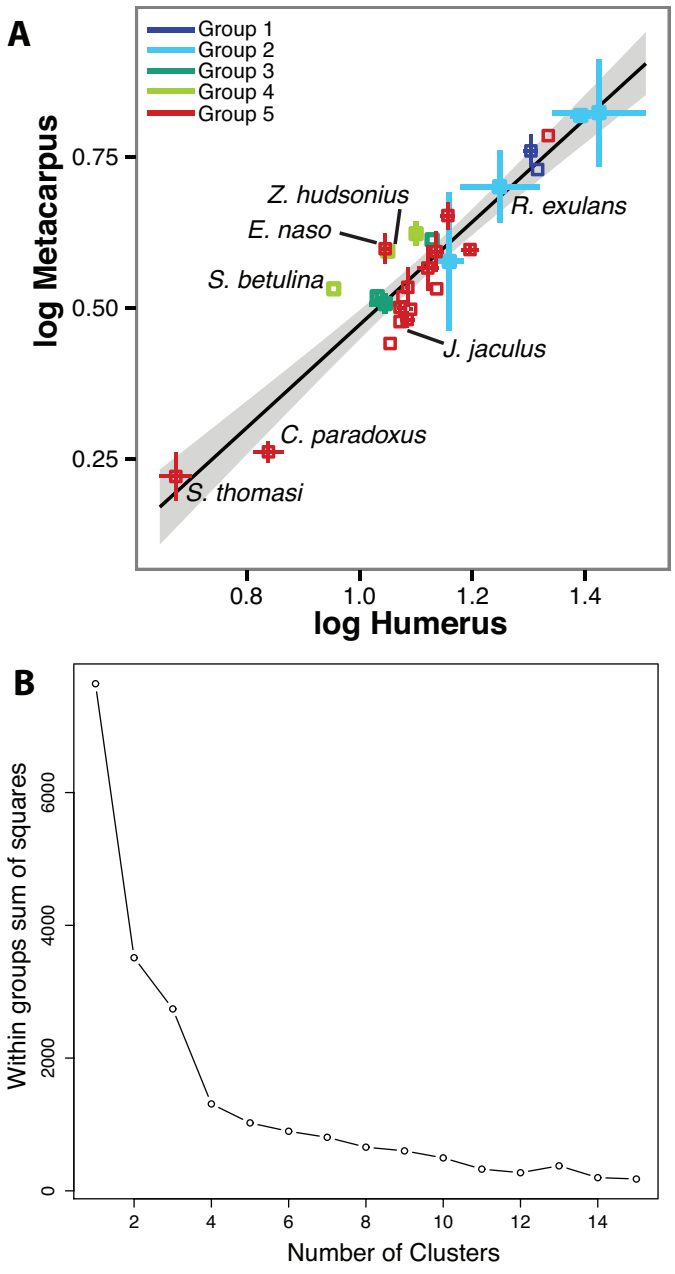

Figure S2. A) The humerus and third metacarpal scale near-isometrically for the ecomorphs identified by our clustering analysis. Log-log plot of the allometric scaling of humerus versus metacarpus. Linear regression: slope=0.853, p=1.322e-13, adjusted R^2=0.878. Colors correspond to ecomorph groups identified in Figure 4B. Grey shading indicates 95% confidence interval, and vertical and horizontal lines represent variation one standard deviation away from the mean. B) Kmeans cluster analysis illustrates the variation in limb proportions by pPC 2, 3, and 4 is best described using five clusters (least change in slope of the line after five clusters). Supports Figure 4.

Table S2. Vertebral lengths (in mm) and ratio of vertebra to previous vertebra

Vertebra	*J jaculus* 1 (mm)	*J jaculus* 2 (mm)	Average (mm)	Ratio n/n-1	*S betulina* (mm)	Ratio n/n-1
T1	1.42	1.57	1.5	1	0.57	1
T2	1.46	1.57	1.52	1.01	0.99	1.74
T3	1.72	1.77	1.75	1.15	1.11	1.11
T4	1.91	1.75	1.83	1.05	1.19	1.08
T5	2.02	1.91	1.97	1.07	1.17	0.98
T6	2.03	2.21	2.12	1.08	1.28	1.09
T7	2.03	2.05	2.04	0.96	1.28	1.01
T8	2.18	2.24	2.21	1.08	1.33	1.03
T9	2.59	2.2	2.4	1.08	1.43	1.08
T10	2.97	2.42	2.7	1.13	1.42	0.99
T11	2.96	2.8	2.88	1.07	1.41	1
T12	2.7	2.32	2.51	0.87	1.5	1.06
(T13)					1.81	1.21
L1	3.24	2.48	2.86	1.14	2.11	1.17
L2	3.25	3.25	3.25	1.14	2.37	1.12
L3	3.99	3.54	3.77	1.16	2.45	1.04
L4	4.29	4.1	4.2	1.11	2.61	1.06
L5	4.29	4.09	4.19	1	2.42	0.93
L6	3.78	3.62	3.7	0.88	2.37	0.98
	J jaculus	*S betulina*				
Average thoracic	2.12	1.27				
Average lumbar	3.59	2.39				
Thoracic /lumbar	0.59	0.53				

Table S3. Hindlimb morphotype classifications for all 51 species of Dipodoidea (summarized by genus) based on metatarsal fusion, number of digits, and limb allometry.

Genus and number of species (sp)	Metatarsal fusion	Number of digits	Locomotion/ Limb Allometry Group (Fig 4B, 5)	Hindlimb morphotype (Fig 2)
Sicistinae				
Sicista (13 sp)	None	5	Quadruped	A
Zapodidae				
Eozapus (1 sp)	None	5	Basal facultative biped	B
Napaeozapus (1 sp)	None	5	Basal facultative biped	B
Zapus (3 sp)	None	5	Basal facultative biped	B
Cardiocraniinae				
Salpingotus (5 sp)	None	3	Basal pygmy biped	C
Salpingotulus (1 sp)	None	3	Basal pygmy biped	C
Cardiocranius (1 sp)	None	5	Basal pygmy biped	D
Euchoreutinae				
Euchoreutes (1 sp)	Partial	5	Basal biped	E
Allactaginae				
Allactaga tetradactyla	Full	4	Derived biped	F
Allactaga (11 sp)	Full	5	Derived biped	G
Pygeretmus (3 sp)	Full	5	Derived biped	G
Dipodinae				
Dipus (1 sp)	Full	3	Derived biped	H
Eremodipus (1 sp)	Full	3	Derived biped	H
Jaculus (3 sp)	Full	3	Derived biped	H
Stylodipus (3 sp)	Full	3	Derived biped	H
Paradipus (3 sp)	Full	3	Derived biped	H

Table S4. Loadings (top) and species scores (bottom) from the phylogenetically-corrected Principal Components Analysis.

	PC1	PC2	PC3	PC4
humerus	-0.927962659	0.356347462	0.105019557	-0.02954119
femur	-0.988392059	0.106862461	-0.062804684	0.087847158
tibia	-0.988676775	-0.061289937	-0.119702004	-0.066582349
metatarsal	-0.905125775	-0.415083981	0.091664625	0.00708643
Allactaga_sibirica	-0.833498727	-10.35657937	-1.06648016	0.880361456
Allactaga_major	-10.39899369	-9.765816326	-1.038119269	0.938893537
Allactaga_bullata	0.702943694	-10.06921166	-1.035308625	1.068422691
Allactaga_elater	3.442629678	-11.33156407	-1.354898378	1.155256564
Allactaga_tetradactyla	2.618551426	-10.76311821	-1.303189771	0.803366754
Jaculus_jaculus	2.428835486	-11.96420373	-1.019793281	0.230172191
Jaculus_blanfordi	-0.740042387	-11.4972549	-0.972631483	0.295664309
Stylodipus_andrewsi	3.452682919	-9.502045937	-1.287213984	1.706944543
Dipus_sagitta	1.587425245	-10.08717732	-1.069563948	0.936617127
Euchoreutes_naso	7.902322257	-10.23825472	0.33567902	-0.138128795
Salpingotus_koslovi	30.28733374	-7.963214022	-1.32157391	0.614203907
Cardiocranius_paradoxus	20.82912937	-7.920307289	-0.749194601	0.977086321
Zapus_hudsonius	16.27731228	-4.411206318	1.593165947	0.608551562
Napaeozapus_insignius	14.28267712	-3.752875384	1.603025726	0.50488667
Sicista_betulina	26.19246792	-0.825554552	1.066899444	0.187256716
Microtus_pennsylvanicus	13.82091084	3.128606717	0.50192181	-0.288222943
Microtus_mexicanus	21.50728549	3.18689337	0.118065083	0.194682335
Microtus_arvalis	24.68966035	2.924844468	1.262136072	-0.309368234
Mesocricetus_brandti	10.13057769	9.039933305	-1.737488265	0.561997069
Mesocricetus_auratus	13.6100043	7.012090742	0.019907347	1.170157765
Peromyscus_leucopus	17.50003803	0.878632183	0.505103325	0.346229223
Peromyscus_polionotus	22.64682191	-0.065587696	1.226285782	0.619878336
Rattus_exulans	10.32243591	1.31378492	0.006553186	0.251760162
Rattus_rattus	4.656035714	2.156644903	-0.057864511	0.432177148
Rattus_norvegicus	2.710245132	2.754811174	0.63694964	-0.017983533
Mus_musculus	21.93967027	0.269771598	1.082439015	0.825438403
Acomys_russatus	17.27645196	3.459604153	-0.634706936	0.54861346
Spalax_ehrenbergi	17.04856928	8.609635305	-0.384676429	0.085116147
Rhizomys_pruinosus	-3.430436863	6.667702878	0.911769789	0.625133264
Anomalurus_beecrofti	-11.81791689	8.941832207	-1.070598583	-0.50411194
Pedetes_capensis	-20.24209142	-4.157693636	-1.701977913	-1.121037362
Thomomys_bottae	8.521456161	4.065084169	1.219275488	0.517551453
Thomomys_umbrinus	11.69965665	3.171629521	1.786969911	1.002643689

Geomys_bursarius	5.07194447	1.995062152	-1.066298917	1.981288997
Dipodomys_ordii	7.538328416	-4.805912605	-0.934676363	0.357178695
Dipodomys_spectabilis	0.608328366	-4.281753454	-1.476500694	-0.185439194
Castor_canadensis	-17.81466575	-1.029094077	2.041696055	-1.054662598
Castor_fiber	-21.87861557	-1.210462642	2.120834544	-1.490736905

Table S5. ANCOVA for allometric scaling regression lines of quadrupeds and most derived of the bipeds.

ANCOVA Models	Lh	Alpha	Beta 2	Beta 3	Beta 4	R^2	s.e. Alpha	s.e. Beta-2	s.e. Beta-3	s.e. Beta-4	Lambda	Pval	Preference for two lines
Leg vs arm: Single line	19.197768	0.338402709	1.067845112	-	-	0.94509387	0.217688	0.056166	-	-	1	3.99E-14	
Leg vs arm: Two lines	36.166265	0.0699296	1.071690265	1.129301469	-0.092365784	0.996520147	0.068786	0.019941	0.288094	0.080001	0.0000001	4.35E-21	4E-08
Metatarsal vs femur: Single line	32.667358	0.056812711	0.792952297	-	-	0.787746896	0.132546	0.08982	-	-	1	1.88E-08	
Metatarsal vs femur: Two lines	40.708114	0.117630554	0.651398062	0.033229927	0.241302717	0.984655062	0.097666	0.074957	0.227384	0.15434	0.0000001	1.3E-15	3E-03

Table S6. Statistical analysis of association of (log) element length with the nodes demonstrating punctuated evolution of the hindlimb.

Punctuation Models	Lh	Alpha	Beta 2	R^2	s.e. Alpha	s.e. Beta-2	Lambda	T-value	d.f.	Pval	Order	Critical Value	Adj Pval
Tibia vs nodes	24.167607	0.904784222	0.066880819	0.762270056	0.06989	0.007325	1.00E-07	9.130487213	24	1.41E-09	1	0.001666667	8.45E-09
Femur vs nodes	21.437858	0.858605521	0.057473085	0.65745966	0.077627	0.008136	1.00E-07	7.064046814	24	1.33E-07	2	0.002	6.63E-07
Metatarsals vs nodes	22.457627	0.680702845	0.051933183	0.293375674	0.155472	0.015807	1	3.285454723	24	0.001560335	3	0.0025	0.006241339
Ulna vs nodes	24.108331	0.96690848	0.030684674	0.164002748	0.131144	0.013587	0.962693417	2.258384782	24	0.016644107	4	0.003333333	0.049932322
Humerus vs nodes	19.292325	0.835001406	0.033308963	0.153010398	0.146063	0.015369	0.91857176	2.167282366	24	0.020179452	5	0.005	0.040358904
Metacarpals vs nodes	19.403949	0.42321027	0.019112974	0.052009299	0.15381	0.016003	0.951986213	1.194336957	24	0.122008513	6	0.01	0.122008513

Supplemental Experimental Procedure. Nexus file for tree constructed of 28 Myomorpha including 23 Dipodoidea.

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CCAGCACCGAGAGGAAAGTGGGTCTGAGTGTTGGTCCCCTCTCTGGGAGACTAGCATTGAATGAA
CAGAAATCTYTATGTGCCRAGACTCCTGCAGATAACGAAGATCTTGCTTGCTTAACCCGAAACAG
CAGCATTCTGAAAGTGAACGAGTGGCTTTCCAGAAGTGATGAGTTGTTAACT------
GATACAAACAACATGAGGGGTAAATCAAATGCTGAAGTAACTGGGGCATTAGAAGTTTTCAATA
AAGTAAGCGGAAATTCCTATTCTTCCAAGAAAATGGGTGTATCAGACACTGATCCTTGTCCTGCT
TTACTGTGTCAAAGTGAGACAGTCTGCTCCAAACC---
AGAGAGTAATATCAAAGATAAAATATTTGGGAAAATCTATC---
ATAAGAAGGCAGGCCTCCCCAACTTGAACCATGTGACTGAAAATCTAATTCCAGGAGCATTTGCT
ACAGAACGACAAATAACACAAGAGCATTCC----------GGTG-------
CACAGATGTCCCAATCAGCCCTACCACCCCGTATGTAGAAGACACCCCCGAGCCGCCCCTGCACGA
CTTCTACTGCAGTAAGCTGCTGGATCTGGTCTTCCTGCTGGACGGCTCCTCCCAGCTGTCYGAGG
CTGAGTTTGAAGTGCTCAAGGCGTTTGTGGTCAGCACAATGGAGAGGCTGGACATCTCGCAGAA
GCGCRTCCGCGTGGCCGTCGTGGAGTATCACGATGGTTCCCACGCCTACATCGAGCTCAAGACCC
GGAAACGGCCCTCAGAGCTGCGACACATTGCCAGCCACGTGAAGTACGCTGGCAGCCAGGAGGCC
TCCACCAGCGAGGTCTTGAAGTACACCCTGTTTCAAATATTCGGCAACATTGATCGGCCAGAAGC
TTCCCGCATCACGCTGCTCCTCACCGCCAGCCAGGAGCCCCCGCGGATGGTCAGGAACCTG---------
GTCCGCTACGTC----------CAGGGCTTG----------


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ATTTATTTCATTTCTATTGGTTGCATGAACAACAAAACATATATTTTCCATTGTACAGAGAAAG
ACTTGGTAG---
ATGTTCCCAAAGATAGATATGGCCATTCCATTGATGTGGCGTATAGTTGTGGGAAAAGTATGGA
TGTTCTCTTTGGAGACCAGTCCTACATGCCTTCTTCCCAAAGAACCACAGAAACATGGGGTAGGG
TAGCTGACTGCCTGCTCCGTGTTTTTCTGGTGGATTTTGAATTTGGGTGTGCTACATCATACATT
CTTCCAGAAC---------
AGCTATCTTTTCATGTCTCTGTTGCCAGAAATGATATCATTTACATTTCAGGAGGACACTCACTT
G---
TGAATATCCANTCTGCTGATCTGTACAGAATAGGGGTTGATCGTCCCCTGGGTAGCCCAGCTGTG
AGTTGCACAATCTTA----------------------------------------------------------------------------------------------------
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        ;
End;

begin mrbayes;
        [This block sets up several different partitions that could be used in the analysis
         of this dataset. Note that Ibalia is selected as the outgroup; the default outgroup
      is the first taxon in the matrix.]
        outgroup Dipodomys_ordii;

        charset A2AB = 1-504;
        charset CNR1 = 505-982;
        charset GHR = 983-1629;
        charset IRBP = 1630-2165;
        charset LCAT = 2166-2545;
        charset BRCA1 = 2546-3196;
        charset vWF = 3197-3729;
        charset ATP7a = 3730-4319;
        charset CREM = 4320-4795;
        charset RAG2 = 4796-5254;
        charset DMP1 = 5255-5741;
        partition Names = 11: A2AB, CNR1, GHR, IRBP, LCAT, BRCA1, vWF, ATP7a, CREM,
RAG2, DMP1;
        set partition=Names;
        lset applyto=(all) nst=6 rates=invgamma;
```

[Unless the parameters are explicitly unlinked, they will be shared across partitions. The unlink command is used to unlink the parameters.]
unlink shape=(all) pinvar=(all) statefreq=(all) revmat=(all);

[ADD A CLOCK RATE]
constraint root = 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 22 23 24 25 26 27 28;
constraint ingroup = 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 20 21 23 24 25 26 27 28;
constraint Dipodidae = 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18 19 23 24 25 26 27 28;
constraint mouse.rat = 1 2;
constraint cardiocraniine = 19 23;
constraint Dipodinae.Allactaginae = 6 7 8 9 10 11 12 13 15 16 17 18 24 25 26 27 28;
constraint Jaculus = 15 16 27;

[constraint <name of constraint> <type of constraint> = <taxa>]
prset topologypr = constraints(root, ingroup, Dipodidae, mouse.rat, cardiocraniine, Dipodinae.Allactaginae, Jaculus);

[node calibrations. The offset exponential distribution has its expectation at the minimum age and mean. That is, the
offsetexponential(300,325) has the minimum 300 and an exponential mean of 325. branch lengths in millions of years]
calibrate root = offsetexponential(55.8, 59); [Calibrations taken from Date-a-Clade]
calibrate Dipodidae = offsetexponential(43, 47); [Calibrations taken from Wu et al 2012, PLoS One]
calibrate mouse.rat = offsetexponential(7.3, 9); [Calibrations taken from Wu et al 2012, PLoS One]
calibrate cardiocraniine = offsetexponential(9, 11); [Calibrations taken from Wu et al 2012, PLoS One]
calibrate Dipodinae.Allactaginae = offsetexponential(10.5, 13); [Calibrations taken from Wu et al 2012, PLoS One]

prset applyto = (all) ratepr = variable;
prset brlenspr = clock: uniform; [need uniform clock when you include fossil species]
prset nodeagepr = calibrated;
prset clockratepr = normal(0.2, 0.02);
[normal(mean, variance)]
prset clockvarpr = igr;
prset Igrvarpr = exponential(0.8);

[The following line allows rates to vary across partitions]
prset ratepr=variable;
mcmcp ngen=10000000 nchains=4 nruns=2 printfreq=1000 samplefreq=1000;

```
end;
```