ELECTRONIC APPENDIX

This is the Electronic Appendix to the article

A Comparative Analysis of the Evolutionary Relationship Between Diet and Enzyme Targeting in Bats, Marsupials and Other Mammals

by

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ONLINE APPENDIX

Appendix table A.

Comparative analysis of AGT subcellular distribution with various character traits.

A) Maximum likelihood analysis using the CONTINUOUS computer program. Log likelihood values (lnL) were calculated for the hypothesis H_1 (i.e. that there is a relationship between the two character traits) and for the null hypothesis H_0 (i.e. that there is no association between the two traits). The likelihood ratio test statistic is calculated as twice the log likelihood difference, and is compared with a chi^2 distribution to test whether the alternative hypothesis fits the data significantly better than the null hypothesis. **B)** Phylogenetically independent contrasts were calculated using the CAIC computer program. Linear regression through the origin was used to statistically analyse the relationship between contrasts. The number of data points (N) included in each analysis varied because not all values were available for all species.

Appendix Table A

a) CONTINUOUS

Traits compared	**N**	**lnL H_0**	**lnL H_1**	**Test statistic**	**P value**	**Correlation**
AGT distribution & diet	77	-209.983	-178.662	62.642	<0.001	0.751
AGT distribution & body mass	77	-238.094	-235.546	5.096	0.024	0.253
AGT distribution & gestation length	70	-136.404	-135.378	2.052	0.152	0.170
AGT distribution & AGT immunoreactivity	38	-88.8125	-88.175	0.128	0.259	-0.182
AGT distribution & AGT catalytic activity	38	-98.755	-98.317	0.876	0.349	-0.159
Diet & body mass	77	-293.096	-291.538	3.116	0.078	0.199

b) CAIC

Traits compared	N	R^2	Slope	F ratio	P value
AGT distribution & diet	77	0.561	1.535	90.618	<0.001
AGT distribution & body mass	77	0.069	0.765	5.291	0.024
AGT distribution & gestation length	70	0.035	0.161	2.420	0.125
AGT distribution & AGT immunoreactivity	38	0.032	-0.228	1.200	0.281
AGT distribution & AGT catalytic activity	38	0.020	-0.228	0.736	0.397
Diet & body mass	77	0.041	0.285	2.998	0.088

Appendix Figure A.

Immuno-gold electron microscopic analysis of AGT distribution in bat livers.

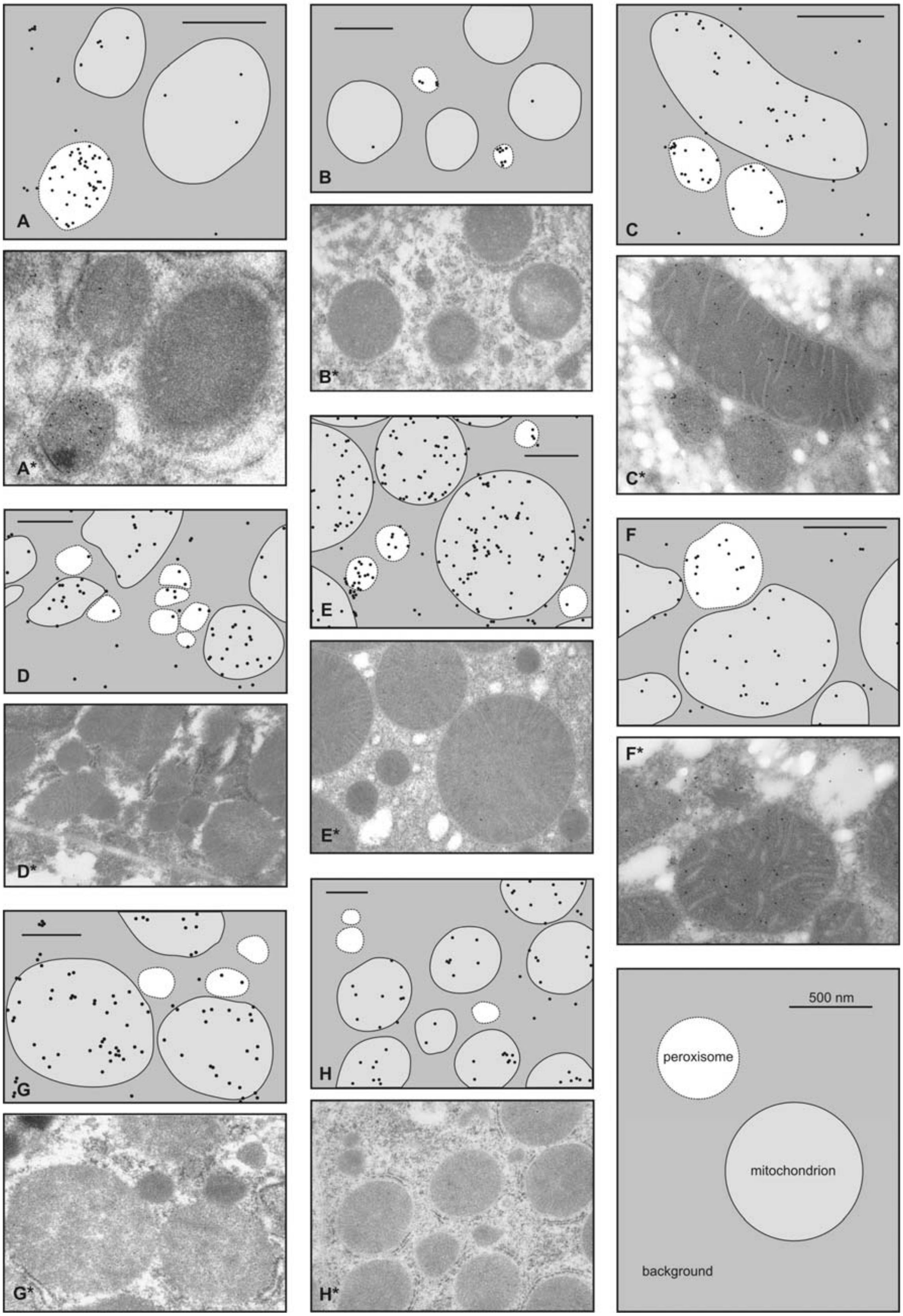
Immunoreactive AGT was determined in various bat livers using the immunogold technique (see Methods section). In order to aid visibility, panels A-H are traces of the immunoelectron micrographs in panels A*-H*, respectively. A, Seba's short-tailed fruit bat (*Carollia perspicillata*); B, Rodriguez fruit bat (*Pteropus rodricensis*); C, serotine bat (*Eptesicus serotinus*); D, noctule bat (*Nyctalus noctula*); E, pipistrelle bat (*Pipistrellus pipistrellus*); F, long-eared bat (*Plecotus auritus*); G, common vampire bat (*Desmodus rotundus*); H, horseshoe bat (*Rhinolophus ferrumequinum*). In the traces, peroxisomes are shaded white and bounded by dashed lines. Mitochondria are shaded light grey and bounded by solid lines. Bars = 500nm. Bats A and B are categorised as peroxisomal, C-F as mitochondrial + peroxisomal, and G and H as mitochondrial.

Appendix Figure B.

Immuno-gold electron microscopic analysis of AGT distribution in miscellaneous mammalian livers.

Immunoreactive AGT was determined in various mammalian livers using the immunogold technique (see Methods section). In order to aid visibility, panels A-F are traces of the immunoelectron micrographs in panels A*-F*, respectively. A, cuscus (*Strigocuscus gymnotis*); B, Przewalski's horse (*Equus przewalskii*); C, dunnarts (*Sminthopsis crassicaudata*); D, slow loris (*Nycticebus coucang*); E, short-nosed echidna (*Tachyglossus aculeatus*); F, oriental small-clawed otter (*Amblonyx cinereus*). In the traces, peroxisomes are shaded white and bounded by dashed lines. Mitochondria are shaded light grey and bounded by solid lines. Bars = 500nm. A and B are categorised as peroxisomal, C and D as mitochondrial + peroxisomal, and E and F as mitochondrial.

Appendix Figure A



Appendix Figure B