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Reporting Summary

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When statistical analyses are reported, confirm that the following items are present in the relevant location (e.g. figure legend, table legend, main

Statistical parameters

text	text, or Methods section).					
n/a	Со	nfirmed				
	\boxtimes	The exact sample size (n) for each experimental group/condition, given as a discrete number and unit of measurement				
	\boxtimes	An indication of whether measurements were taken from distinct samples or whether the same sample was measured repeatedly				
	\boxtimes	The statistical test(s) used AND whether they are one- or two-sided Only common tests should be described solely by name; describe more complex techniques in the Methods section.				
\boxtimes		A description of all covariates tested				
	\boxtimes	A description of any assumptions or corrections, such as tests of normality and adjustment for multiple comparisons				
	\boxtimes	A full description of the statistics including <u>central tendency</u> (e.g. means) or other basic estimates (e.g. regression coefficient) AND <u>variation</u> (e.g. standard deviation) or associated <u>estimates of uncertainty</u> (e.g. confidence intervals)				
\boxtimes		For null hypothesis testing, the test statistic (e.g. <i>F</i> , <i>t</i> , <i>r</i>) with confidence intervals, effect sizes, degrees of freedom and <i>P</i> value noted Give <i>P</i> values as exact values whenever suitable.				
\boxtimes		For Bayesian analysis, information on the choice of priors and Markov chain Monte Carlo settings				
\boxtimes		For hierarchical and complex designs, identification of the appropriate level for tests and full reporting of outcomes				
	\boxtimes	Estimates of effect sizes (e.g. Cohen's d, Pearson's r), indicating how they were calculated				
	\boxtimes	Clearly defined error bars State explicitly what error bars represent (e.g. SD, SE, CI)				

Our web collection on statistics for biologists may be useful.

Software and code

Policy information al	pout <u>availability of computer code</u>
Data collection	RNA quality was assessed using Fragment Analyzer (Advanced Analytical). Raw reads were obtained using Illumina's HiSeq 2500 control software.
Data analysis	We used GSNAP (22-10-2014) to map the RNA-seq reads against the reference genomes. We used HTSeq (0.6.1) to generate read counts from these alignments. We used EdgeR (3.14.0) to perform normalization and generate the expression tables. We used DESeq2 (1.12.4) to create variance stabilizing transformed counts and to do differential gene expression analyses between adjacent time points. The alignment files were manipulated using samtools (0.1.18) and Bedtools (2.18), and general alignment statistics were created using Picard (1.86). PCAs were done using FactoMineR (1.34). We identified genes with significant temporal changes during organ development using maSigPro (1.44.0). We mapped developmental stages across species using the R package dtw (1.18-1). We identified the most common profiles (clusters) during development using mFuzz (2.32.0). We compared developmental trajectories between species using GPClust. Functional enrichments were done using the R implementation of WebGestalt (0.0.5). All statistical analyses and plots were done in R (3.3.2) as implemented in Rstudio (1.0.136). Plots were created using the R packages ggplot2 (2.2.1), gridExtra (2.2.1), reshape2 (1.4.2), plyr (1.8.4), and factoextra (1.0.4).

For manuscripts utilizing custom algorithms or software that are central to the research but not yet described in published literature, software must be made available to editors/reviewers upon request. We strongly encourage code deposition in a community repository (e.g. GitHub). See the Nature Research guidelines for submitting code & software for further information.

Data

Policy information about availability of data

All manuscripts must include a <u>data availability statement</u>. This statement should provide the following information, where applicable:

- Accession codes, unique identifiers, or web links for publicly available datasets
- A list of figures that have associated raw data
- A description of any restrictions on data availability

Raw and processed RNA-seq data are available from ArrayExpress: E-MTAB-6769 (chicken), E-MTAB-6782 (rabbit) E-MTAB-6798 (mouse), E-MTAB-6811 (rat), E-MTAB-6813 (rhesus), E-MTAB-6814 (human) and E-MTAB-6833 (opossum). We also created a publicly available data resource (evodevoapp.kaessmannlab.org), where the profiles of individual genes can be easily visualized and the expression tables can be downloaded.

Field-specific reporting

Please select the best fit for your research. If you are not sure, read the appropriate sections before making your selection.

🔀 Life sciences 👘 Behavioural & social sciences 👘 Ecological, evolutionary & environmental sciences

For a reference copy of the document with all sections, see nature.com/authors/policies/ReportingSummary-flat.pdf

Life sciences study design

All studies must disclose on these points even when the disclosure is negative.

Sample size	Our aim was to study gene expression profiles for 7 major organs from early organogenesis to adulthood in human, mouse, rat, rabbit, rhesus macaque, opossum and chicken. We started sampling as early as it was possible to dissect and isolate the organs and then sampled prenatal development at regular intervals. Postnatally we sampled around the major developmental milestones. The time points sampled were chosen to cover the most important periods of organ development and varied from 9 time points in chicken to 23 in human. We aimed for 4 biological replicates (2 males and 2 females) for somatic organs and 2 replicates for the gonads. Fewer replicates were available for the primates. The number of replicates were chosen based on field standards for differential gene expression analysis; no statistical methods were used to pre-determine sample size. This resulted in 1,893 samples (full details in Supplementary Tables 1 and 2). This dataset provides a detailed, quantitative description of gene expression throughout the development of the cerebrum, cerebellum, heart, kidney, liver, ovary and testis for 6 mammals and a bird.
Data exclusions	We are making available a small number of libraries that were not used in this study because they had a correlation with their biological replicates lower than 0.90. The decision to exclude these libraries was made before performing the analyses described in the manuscript. We are providing these libraries because they were used in other projects from our group (still to be published) and are therefore part of the evo- devo resource. These libraries are clearly marked in Supplementary Table 2.
Replication	We generated biological replicates for the stages and organs sampled in all species. We aimed for 4 biological replicates (2 males and 2 females) for somatic organs (2 for primates) and 2 replicates for the gonads. The analyses described in the manuscript take into consideration the information from the biological replicates. We used PCA and hierarchical clustering to identify and exclude outlier libraries (e.g caused by low RNA quality). We also excluded libraries that showed a Spearman's correlation coefficient with its biological replicates lower than 0.9.
Randomization	All comparisons in this work are based on 3 biological variables: species, organ and developmental stage. Generally, randomization does not apply. When sequencing the RNA-seq libraries (they were multiplexed in sets of 6 or 8) we mixed samples from different organs, stages and species. Full randomization was not possible because the samples arrived at different times and were processed based on their date of reception.
Blinding	Blinding was not relevant to our study. Both data collection and analyses required an understanding of the nature of the sample being collected/analyzed (i.e. species, organ, developmental stage).

Reporting for specific materials, systems and methods

Materials & experimental systems

n/a Involved in the study

 Involved in the study

Methods

- n/a Involved in the study
- ChIP-seq
- MRI-based neuroimaging

Animals and other organisms

Policy information about studies involving animals; ARRIVE guidelines recommended for reporting animal research

Laboratory animals	All mouse (Mus musculus) samples are from the strain CD-1 (RjOrl:SWISS); all rat (Rattus norvegicus) samples are from the outbred strain Holtzman SD; all rabbit (Oryctolagus cuniculus) samples are from the outbred New Zealand breed; all chicken (Gallus gallus) samples are from the red junglefowl, the progenitor of domestic chicken. The other species used in this study were the gray short-tailed opossum (Monodelphis domestica) and rhesus macaque (Macaca mulatta). We sampled males and females in each species.
Wild animals	The study did not involve wild animals.
Field-collected samples	The study did not involve samples collected from the field.

Human research participants

Policy information about <u>studies involving human research participants</u>

Population characteristics	We sampled organs (i.e. forebrain/cerebrum, hindbrain/cerebellum, heart, kidney, liver, ovary and testis) from both males and females, starting at 4 weeks post conception and ending at 60 years of age. The organ, developmental stage and sex of each sample is described in Supplementary Table 2.
Recruitment	There was no direct recruitment for this work. The human prenatal samples were provided by the MRC-Wellcome Trust Human Developmental Biology Resource (HDBR) and were derived from elective abortions with normal karyotypes. The tissue donations were made entirely voluntarily by women undergoing termination of pregnancy. Donors were asked to give explicit written consent for the fetal material to be collected, and only after they had been counselled about the termination of their pregnancy. The human postnatal samples were retrieved from the NICHD Brain and Tissue Bank for Developmental Disorders at the University of Maryland (USA) and from the Chinese Brain Bank Center (CBBC) in Wuhan (China). They originated from individuals with diverse causes of death that, given the information available, was not associated with the organ sampled. Written consent for the use of human tissues for research was obtained from all donors or their next of kin by the respective tissue banks. We are not aware of any potential self-selection biases (or other) that could have affected this work.