

CLAIMS

1. A computer-implemented method of establishing an epigenetic clock for an avian species, the method comprising
 - 5 (a.) identifying and determining the methylation levels of specific CpG sites within the genomic DNA obtained from a plurality of different biological sample materials deriving from the avian species and representing specific time points within the chronological lifespan of this avian species,
 - (b.) excluding all CpG sites associated with single nucleotide polymorphisms from the CpG sites identified in step (a.),
 - 10 (c.) excluding all CpG sites located on the sex chromosomes from the CpG sites obtained in step (b.),
 - (d.) performing a tissue-specific normalization step for the CpG sites obtained in step (c.), and
 - (e.) correlating the CpG methylation levels of the CpG sites obtained in step (d.) with chronological age using a penalized regression model,
 - 15 wherein the specific CpG sites within the genomic DNA of the avian species are distributed within low methylated regions (LMRs) in the genome of the avian species.
2. The method according to claim 1, wherein the plurality of different biological sample materials deriving from the avian species and representing specific time points within the chronological lifespan
 - 20 of this avian species includes material selected from the group consisting of body fluids, excremental material, tissue material and feather material.
3. The method according to claim 1, wherein plurality of different biological sample materials deriving from the avian species and representing specific time points within the chronological lifespan
 - 25 of this avian species includes at least four different tissues.
4. The method according to any one of the preceding claims, wherein the plurality of different biological sample materials deriving from the avian species and representing specific time points within the chronological lifespan of this avian species consists of tissue material selected from muscle
 - 30 tissue, gut tissue, organ tissue and skin tissue.
5. The method according to any one of the preceding claims, wherein the plurality of different biological sample materials deriving from the avian species and representing specific time points within the chronological lifespan of this avian species includes breast tissue, spleen tissue, ileum
 - 35 tissue and jejunum tissue.
6. The method according to any one of the preceding claims, wherein the plurality of different biological sample materials deriving from the avian species and representing specific time points within the chronological lifespan of this avian species is selected to represent ages ranging between
 - 40 3 days and 63 days.

7. The method according to any one of the preceding claims, wherein step (a) involves a whole-genome bisulfite sequencing process.
- 5 8. The method according to any one of the preceding claims, wherein the specific CpG sites within the genomic DNA of the avian species are distributed over the whole genome of the avian species and were restricted to a strand-specific coverage of at least 10.
9. The method according to claim 8, wherein the tissue-specific normalization step is performed
10 by computing for every CpG the average value over all samples from the same tissue and subtracting this value from the value of this CpG.
10. The method according to claim 1, wherein the specific CpG sites within the genomic DNA of the avian species were restricted to a strand-specific coverage of at least greater than 5.
- 15 11. The method according to claim 1, wherein the tissue-specific normalization step is performed by computing for every LMR the average value over all samples from the same tissue and subtracting this value from the value of this LMR.
- 20 12. A computer program loaded into a memory of a computer, implementing the method of any one of claims 1 to 9.
13. A tangible, computer-readable medium comprising a computer-readable code that, when executed by a computer, causes the computer to perform operations comprising:
- 25 (a.) receiving information corresponding to the methylation levels of specific CpG sites within the genomic DNA of the avian species obtained from a plurality of different biological sample materials representing specific time points within the chronological lifespan of the avian species,
- (b.) receiving information corresponding to all CpG sites associated with single nucleotide polymorphisms, and excluding same from the CpG sites of step (a.),
- 30 (c.) receiving information corresponding to all CpG sites from the sex chromosomes and excluding same from the CpG sites of step (b.),
- (d.) performing a tissue-specific normalization step for the CpG sites of step (c.), and
- (e.) correlating the CpG methylation levels of the CpG sites of step (d.) with chronological age using a penalized regression model,
- 35 wherein the specific CpG sites within the genomic DNA of the avian species are distributed within low methylated regions (LMRs) in the genome of the avian species.