Data in Brief 30 (2020) 105640

Contents lists available at ScienceDirect

Data in Brief

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Data Article

Karyological and bioinformatic data on the common chameleon *Chamaeleo chamaeleon*

Marwa Sidhom[a], Khaled Said[a], Noureddine Chatti[a], Fabio M. Guarino[b,*], Gaetano Odierna[b], Agnese Petraccioli[b], Orfeo Picariello[b], Marcello Mezzasalma[b,c]

[a] *Laboratoire de Génétique, Biodiversité et Valorisation des Bioressources, Institut Supérieur de Biotechnologie de Monastir, Université de Monastir, Avenue Taher Hadded (B.P 74) Monastir 5000, Tunisie*
[b] *Dipartimento di Biologia, Università di Napoli Federico II, Via Cinthia 26, 80126 Napoli, Italy*
[c] *Department of Life Sciences, Natural History Museum, Cromwell Road, London, SW7 5BD, UK*

ARTICLE INFO

Article history:
Received 6 March 2020
Revised 9 April 2020
Accepted 24 April 2020
Available online 30 April 2020

Keywords:
Karyotype
C-banding
DNA
Chameleons
Reptiles

ABSTRACT

The data presented in this paper stand as supplementary information of the associated article "Karyological characterization of the common chameleon (*Chamaeleo chamaeleon*) provides insights on the evolution and diversification of sex chromosomes in Chamaeleonidae" [1]. This work provides (i) raw experimental data on the karyology of the common chameleon *Chamaeleo chamaeleon* and (ii) the results of bioinformatic analysis on sex-specific and repeated DNA sequences found in the same species. The karyological information here presented includes traditional staining method (Giemsa staining) and sequential C-banding + fluorochromes performed on Tunisian samples of the species. The sequence data include the alignments of the isolated DNA sequences with homologous sequences found in squamate Short Read Archives (SRAs) and the results of searches in public nucleic acid databases.

* Corresponding author: Fabio M. Guarino.
E-mail address: fabio.guarino@unina.it (F.M. Guarino).

https://doi.org/10.1016/j.dib.2020.105640

Specifications Table

Subject	Agricultural and Biological Sciences
Specific subject area	Animal Science and Zoology Ecology, Evolution, Behaviour and Systematics
Type of data	Image Figure
How data were acquired	Karyological data were acquired using different staining/banding techniques (Giemsa staining, sequential C-banding + fluorochromes) followed by observations with optical (Giemsa staining) and epifluorescence (C-banding + fluorochromes) microscope. Molecular data were acquired by sequencing the amplicons obtained using the primers reported in [2] from DNA of a male of *C. chameleo*. The obtained sequences were manually edited with Chromas Lite 2.1.1. (Technelysium Pty Ltd, South Brisbane, AU) and assembled with BioEdit 7.2.5. [3]
Data format	Raw Analyzed .tif images
Parameters for data collection	Metaphase plates were searched at 10x and 20x magnification and recorded at 100x magnification either with optical or epifluorescence microscopy. Observations in epifluorescence were carried out using filter cubes for Chromomycin A^3 (CMA) and DAPI (excitation/emission wavelength, 445 nm /575 nm and 358 nm/ 461 nm, respectively).
Description of data collection	Data were collected through optical (Giemsa staining) and epifluorescence (C-banding + fluorochromes) microscope. Collection of molecular data was performed by search for identity of the obtained sequences [1] using BLAST Short Read Sequences (SRA) Archive of male 4 of *C. calyptratus* (Accession number SRX3547644) [2]. Alignments among C.Cham-RADseq of *C. chamaeleon* and SRA sequences of *C. calyptratus* were performed using BioEdit [3]
Data source location	Latitude and longitude (and GPS coordinates) for collected samples: Agareb (Tunisia) 34°25'60" N, 10°10'60" E (32 S 608722.99 m E, 3810839.52 m N); Tourief (Tunisia) 36°20'18 N, 8°35'9" E (32 S 462833.91 m E, 4021866.03 m N); Zéramdine (Tunisia) 35°34'16" N, 10°44'6" E (32 S 631429.31 m E, 3911883.93 m N)
Data accessibility	Raw karyological data can be found with this article. Sequence data can be found as reported in [1].
Related research article	Sidhom M., Khaled S., Chatti N., Guarino F.M., Odierna G., Petraccioli A., Picariello O., Mezzasalma M. Karyological characterization of the common chameleon (*Chamaeleo chamaeleon*) provides insights on the evolution and diversification of sex chromosomes in Chamaeleonidae. Zoology. In press https://doi/10.1016/j.zool.2019.125738

Value of the Data

- The karyological data presented here show undifferentiated karyotypes, in terms of chromosome number, morphology and distribution of heterochromatin, between different sexes of *C. chamaeleon*, indicating that sex chromosomes are at an early evolutionary stage.
- Cytogeneticists as well as evolutionary and molecular biologists can benefit from these data.
- Further comparative karyological analyses on other species and genera of Chamaeleonidae can highlight differences in chromosome number/morphology, heterochromatin content and distribution, and distinct sex determination systems.
- The alignments with the newly generated DNA sequences, previously isolated in *C. calyptratus*, show their conservation in *C. chamaeleon.*
- Comparative molecular and bioinformatic analyses on the isolated DNA sequences will add information about their conservation and distribution in other chameleon, squamate and vertebrate taxa.

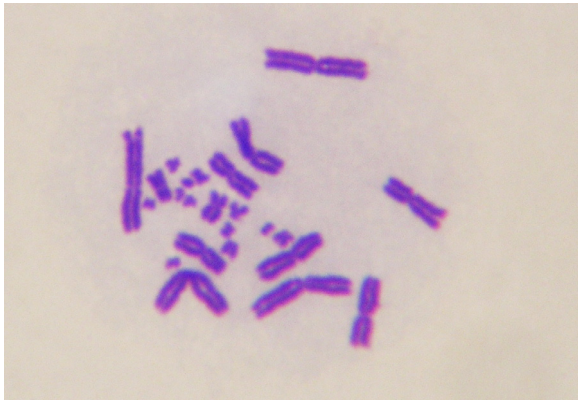

Fig. 1. Metaphase plate of a male specimen of *C. chamaeleon* from Agareb (Tunisia) stained with 5% Giemsa solution.

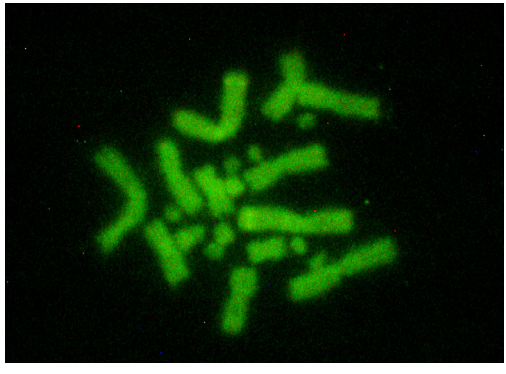

Fig. 2. Metaphase plate of a female specimen of *C. chamaeleon* from Tourief (Tunisia) with C-banding + CMA3.

1. Data Description

The data presented in this paper include the supplementary information of a thorough karyological characterization, molecular and bioinformatic analyses on Tunisian samples of the common chameleon *C. chamaeleon* described in the associated paper [1]. Figure 1 shows a metaphase plate with 2n=24 chromosomes stained with Giemsa solution of a male common chameleon specimen. Figure 2 and 3 represent a sequential C-banding + CMA3 (Fig. 2) and + DAPI (Fig. 3), performed on a metaphase plate from a female specimen of the common chameleon. Figure 4 shows the distribution of best hits from query of the Cham 57 sequence isolated in [1] on SRA (Short Read sequence Archive) sequences of males 1 - 8 of *C. calyptratus* reported in [2]. Figure 5 shows the alignment with C.cham-RADseq (Restriction site-Associated sequence)_M2 sequence isolated in [1] and homologous sequences found in squamate SRAs [2]. Figure 6 shows the alignment with C.cham-RADseq_M3 sequence isolated in [1] and homologous sequences found in squamate SRAs [2]. Figure 7 shows the alignment with C.cham-RADseq_M12 sequence isolated in [1] and homologous sequences found in squamate SRAs [2]. Figure 8 shows the alignment with C.cham-RADseq_M13 sequence isolated in [1] and homologous sequences found in squamate SRAs [2].

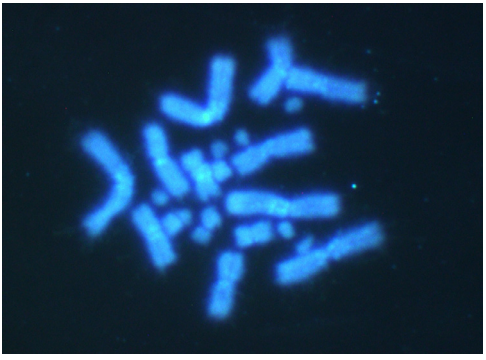

Fig. 3. Metaphase plate of a female specimen of *C. chamaeleon* from Zéramdine (Tunisia) with C-banding + DAPI.

Fig. 4. Distribution of best blast hits of Cham 57 sequence isolated in [1] on SRA sequences of males 1 - 8 of *C. calyptratus* (Accession numbers SRX3547644 - SRX3547651) [3].

2. Experimental Design, Materials, and Methods

We performed different chromosome staining techniques and banding methods to determine the number and morphology of chromosomes and to characterize heterochromatin distribution of the study specimens of the common chameleon. Chromosomes were obtained from cell suspensions using the air-drying method [4]. Specimens were injected with 0.01ml/ g body weight of a 0.1% vinblastine solution (Sigma), and after 2 h they were deeply anaesthetized by exposure to profound exposition to ethyl ether vapours. Chromosomes were obtained from bone marrow using the air-drying method, namely: 5 ml of a hypotonic solution of KCl 0.075 M were injected through medullar canal of femurs and cells collected in a test tube. After centrifugation at 1000 rpm, cells were fixed in methanol-acetic solution (3:1) and dropped (20 microliter) on a slide. Chromosomes were stained with conventional technique (5% Giemsa solution at pH 7 for 10 min.) and with sequential C-banding + CMA+DAPI following [5-7]. In brief, chromosomes were incubated in $BA(OH)_2$ at 40°C (2 min.), washed first in HCl 0.2N and then in distilled water, then incubated in 2xSSC (Sodium Saline Citrate) for 15 min. After washing in distilled water, chromosomes were stained for 20 min in CMA (50 μm/ml in McIlvaine buffer pH7), then washed

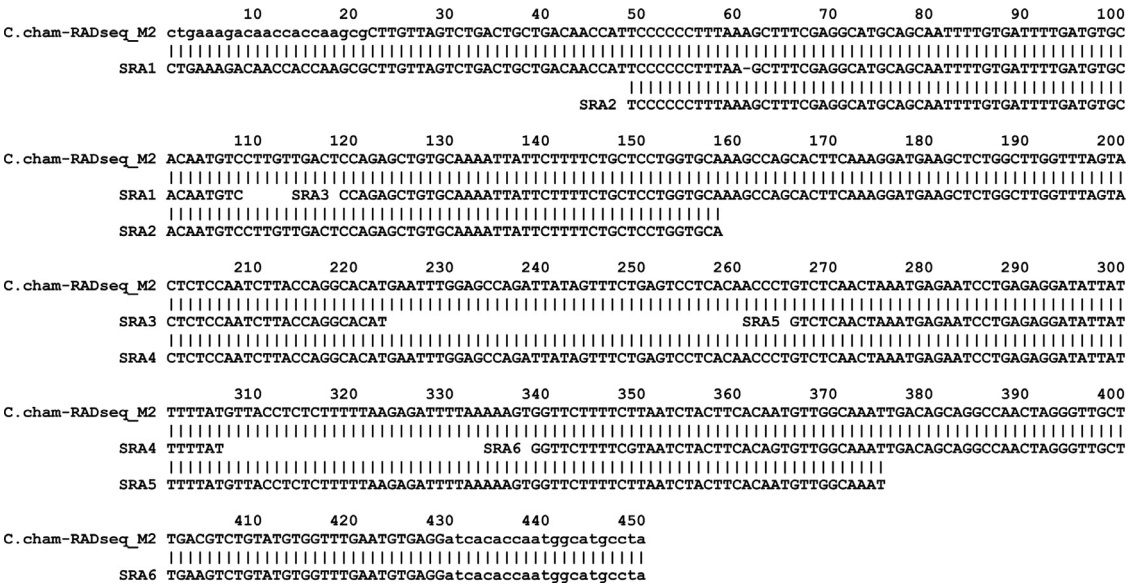

Fig. 5. Alignment of Short Read Archive (SRA 1-6) of RAD seq of *C. calyptratus*: male 4 (Accession number SRX3547644) [3] versus the sequence C.cham-RADseq_M2 of male (Cham57) of *C. chamaeleon* (99% identity) isolated in [1]. Primer pairs are indicated in lower cases.

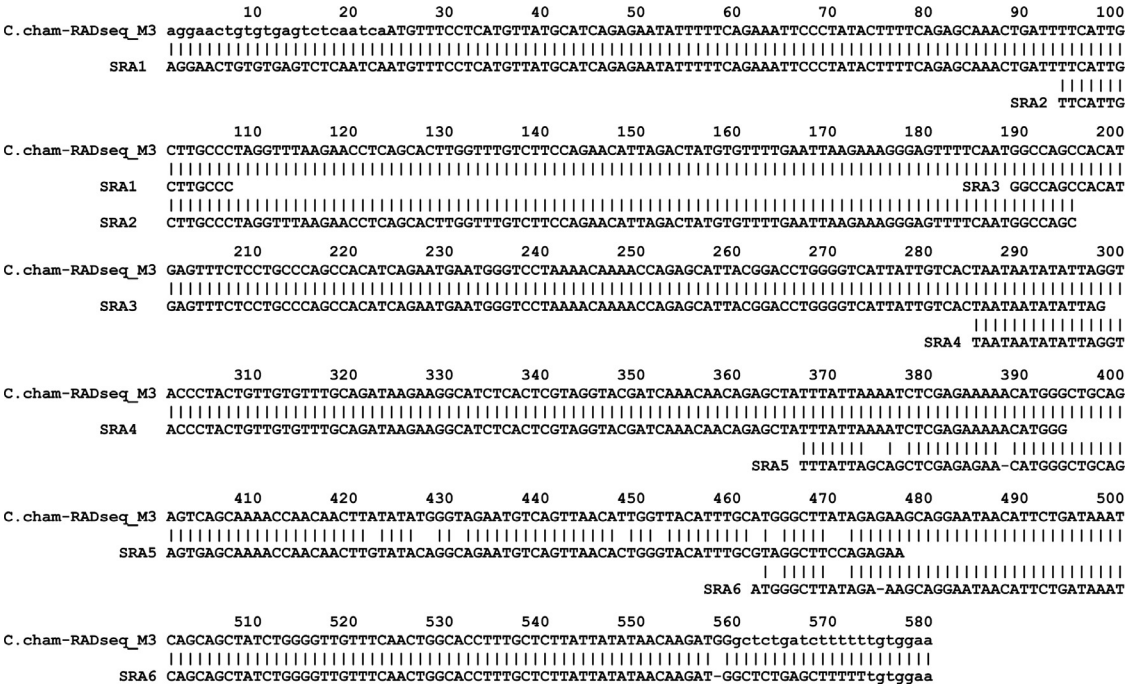

Fig. 6. Alignment of Short Read Archive (SRA 1-6) of RAD seq of *C. calyptratus*: male 4 (Accession number SRX3547644) [3] versus the sequence C.cham- RADseq_M3 of the male (Cham57) of *C. chamaeleon* (99.9% identity) isolated in [1]. Primer pairs are indicated in lower cases.

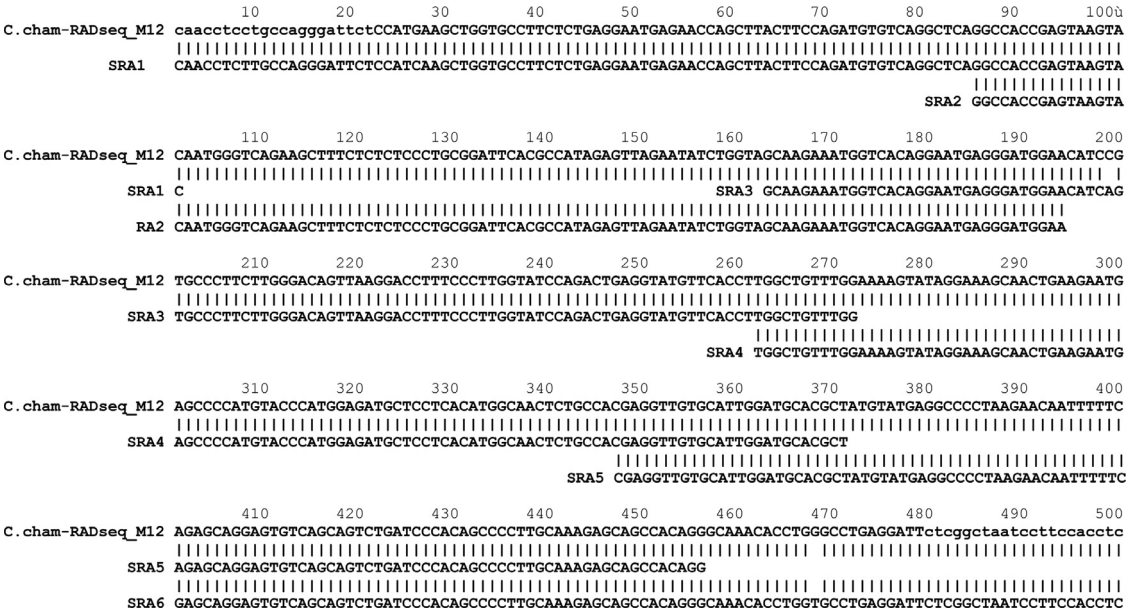

Fig. 7. Alignment of Short Read Archive (SRA 1-6) of RAD seq of *C. calyptratus*: male 4 (Accession number SRX3547644) [3] versus the sequence C.cham-RADseq_M12 of male (Cham57) of C. chamaeleon (identity 99.9%) isolated in [1]. Primer pairs are indicated in lower cases.

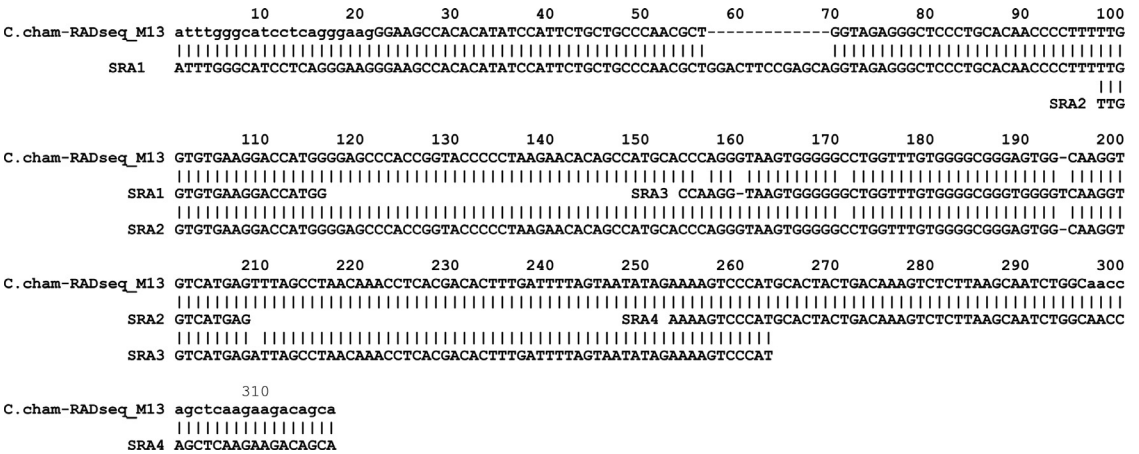

Fig. 8. Alignment of Short Read Archive (SRA 1-4) of RAD seq of *C. calyptratus*: male 4 (Accession number SRX3547644) [3] versus the sequence RADseq Cham_M13of male (Cham57) of *C. chamaeleon* isolated in [1] (identity 95.3%). Primer pairs are indicated in lower cases.

in McIlvaine buffer and stained with DAPI (0.5 μm/ml in McIlvaine buffer) for 10 minutes. After washing in McIlvaine buffer, slides were mounted with antifade solution (80% glycerine + 0,2% of DABCO, DiAzoBiCicloOctane). Determination of the chromosome number, karyotype reconstruction and chromosome classification were performed according to Levan et al. [8].

Metaphase plates were searched using the objectives 10 or 20x with optical (for Giemsa staining) and fluorescence microscope (for sequential C-banding) and recorded with objective 100x.

The DNA sequences isolated in [1] were blasted in BLAST Short Read Sequences (SRA) Archive of male 4 of *C. calyptratus* (Accession number SRX3547644) [2]. Alignment among the DNA sequences isolated in [1] and SRA sequences of *C. calyptratus* were performed using BioEdit [3].

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships which have, or could be perceived to have, influenced the work reported in this article.

Acknowledgments

We thank the Tunisian Ministry of Higher Education and Scientific Research, Tunisia and University of Monastir for giving permission to catch the specimens and for granting research funding at the Biology Department of the University of Naples Federico II.

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