Characterization of the complete chloroplast genome of a Peruvian landrace of *Capsicum chinense* Jacq. (Solanaceae), arnaucho chili pepper

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MITOGENOME ANNOUNCEMENT

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Peru harbors the largest morphological diversity of cultivated chili peppers worldwide (Meckelmann et al. 2013), and they play a crucial role in the Peruvian cuisine and cultural traditions (Morales-Soriano et al. 2018). Arnaucho chili pepper is a landrace cultivated by small farmers in a restricted area named ‘Valle de Supe’ (around 200 km northern Lima) and is an important representative in the gastronomy of Lima region. This landrace possesses an annular constriction of the calyx, two flowers per axil, red ripe fruit color, triangular fruit shape, pointed fruit apex shape, greenish-yellow corolla (Aliaga et al. 2019). Even though NGS techniques are being employed to study *Capsicum* L. spp. genomes, very little is known about the genomics of Peruvian chili pepper. In addition, knowledge about Peruvian *Capsicum chinense* Jacquin 1777 phylogenetic relationships is scarce. Therefore, we here report the first complete chloroplast genome (cp) of a Peruvian chili pepper landrace by next-generation sequencing technology. Moreover, a phylogenetic tree of this species and its relatives is presented.

We collected young fresh leaves of arnaucho chili pepper from the Lima region (−10.8099889, −77.6953950). This specimen was deposited at the Germplasm Bank of INIA (https://www.gob.pe/inia, drgb@inia.gob.pe) under the voucher number PER1002642. Total genomic DNA was extracted by CTAB method (Doyle and Doyle 1990). Pair-end clean reads were obtained by PE 150 library and the Illumina HiSeq 2500 platform. Adapters and low-quality reads were removed using Trim Galore (Martin 2011). We assembled the chloroplast genome using the GetOrganelle v1.7.2 pipeline (Jin et al 2020). Chloroplast genome was annotated with GeSeq in CHLOROBOX web service (Tillich et al 2017).

The total length of the chloroplast genome is 156,931 bp, which is 1,635 bp longer than one of the most economically important species in the Solanaceae family, potato (*Solanum tuberosum*). This cp genome presents a typical quadripartite structure, containing 87,325 bp as large single copy (LSC) region and 17,912 bp as small single-copy (SSC) region, separated by two inverted repeats (IR) regions (25,847 bp); and the percentage of GC content was 37.71%. Arnaucho chili pepper chloroplast genome possesses 133 genes that consists of 86 protein-coding genes, 37 tRNA, eight rRNA, and two pseudogenes. Phylogenetic analysis revealed that this Peruvian chili pepper landrace is closely related to the undomesticated species *C. galapagoense*; all belong to the Capsicaceae tribe.
aligned by MAFFT v7.475 (Katoh and Standley 2013). Then, we used GTR + GAMMA model of evolution to obtain the best-scoring ML tree, and then 1,000 nonparametric bootstrap inferences were performed with RAxML v8.2.11 (Stamatakis 2014). The phylogenomic analyses were consistent with a previous study that also employed plastome sequences (D’Agostino et al. 2018; Sebastin et al. 2019). Interestingly, ML phylogenetic analysis showed that arnacucho chili pepper is sister to \textit{C. galapagoense} and sister to them is a clade containing \textit{C. chinense} and \textit{C. eximium} Hunziker 1950 (Figure 1). This chili pepper landrace is considered a \textit{C. chinense} species by many authors based mainly on morphological characters (Aliaga et al. 2019). However, its current taxonomy is questioned based on the present work. Carrizo García et al. (2016) showed that \textit{C. galapagoense} is nested among the closely related \textit{C. frutescens}, \textit{C. chinense}, and \textit{C. annuum} Linnaeus 1753 as demonstrated also by Ince et al. (2010). In a more recent study, Shiragaki et al. (2020) indicated that the \textit{C. chinense} clade might be divided into two groups. Similarly, Tripodi et al. (2021) demonstrated that \textit{C. chinense} species grouped in two clusters.

To our best knowledge, this is the first report of a cp genome of a Peruvian chili pepper landrace. However, we consider that further studies using additional collections of \textit{Capsicum} from a wider geographic area and examination of relevant type material are needed to provide a better understanding of taxonomic variation and nomenclature in the \textit{Capsicum} clade. A similar process was followed by Arbizu, Ellison, et al. (2016); Arbizu, Simon, et al. (2016) and Martinez-Flores et al. (2016) solving the species boundaries in another problematical group in \textit{Daucus}. Our next step is to continue developing additional molecular tools for the arnacucho chili pepper that may shed light on elucidating its evolutionary history and promoting its adequate sustainable management, conservation and modern breeding.

**Ethical approval**

Research and collection of plant material was conducted according to the guidelines provided by INIA. Permission was granted by grower of “arnaucho” chili pepper to carry out research on this landrace.

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**Disclosure statement**

No potential conflict of interest was reported by the author(s).

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Author contributions

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Data availability statement

The genome sequence data that supports this study is openly available in Genbank of NCBI under the accession number MZ379791 (https://www.ncbi.nlm.nih.gov/nuccore/MZ379791.1/). The associated Bioproject, Biosample and SRA numbers are PRJNA739476, SAMN19789523, and SRR14868519, respectively.

References

Aliaga C. J., Portalatino E., Obregón K., Rodríguez A., Jimenez J. 2019. Presencia del "aji nativo supano" (Capsicum chinense Jacq.) en el valle de Supe, Perú. Peruv Agric Res. 1(2):58–63.
Arbizu C.I., Ellison S.L., Senalik D., Simon P.W., Spooner D.M. 2016. Genotyping-by-sequencing provides the discriminating power to investigate the subspecies of Daucus carota (Apiaceae). BMC Evol. Biol. 16(1):1–16.
Arbizu C.I., Simon P.W., Martinez-Flores F., Ruess H., Crespo M.B., Spooner D.M. 2016. Integrated molecular and morphological studies of the Daucus guttatus complex (Apiaceae). Syst Bot. 41(2):479–492.
Carrizo García C., Barfuss MH., Sehr EM., Barboza GE., Samuel R., Moscone EA., Ehrendorfer F. 2016. Phylogenetic relationships, diversification and expansion of chili peppers (Capsicum, Solanaceae). Ann Bot. 118(1):35–51.
D’Agostino N., Tamburino R., Cantarella C., De Carluccio V., Sannino L., Cozzolino S., Cardi T., Scotti N. 2018. The complete plastome sequences of eleven capscium genotypes: insights into DNA variation and molecular evolution. Genes. 9(10):503.
Doyle JJ., Doyle JL. 1990. Isolation of plant DNA from fresh tissue. Focus. 12(13):39–40.
Ince A.G., Karaca M., Onus AN. 2010. Genetic relationships within and between capscium species. Biochem Genet. 48(1–2):83–95.
Jin JJ., Yu WB., Yang JB., Song Y., Depamphilis CW., Yi TS., Li DZ. 2020. GetOrganelle: a fast and versatile toolkit for accurate de novo assembly of organelle genomes. Genome Biol. 21(1):1–31.
Katoh K., Standley DM. 2013. MAFFT multiple sequence alignment software version 7: improvements in performance and usability. Mol Biol. Evol. 30(4):772–780.
Martin M. 2011. Cutadapt removes adapter sequences from high-throughput sequencing reads. EMBnet J. 17(1):10–12.
Martínez-Flores F., Arbizu C.I., Reitsma K., Juan A., Simon P.W., Spooner D.M., Crespo M.B. 2016. Lectotypic designation for seven species names in the Daucus guttatus complex (Apiaceae) from the central and eastern Mediterranean basin. Systematic Bot. 41(2):464–478.
Meckelmann SW., Riegel DW., van Zonneveld M., Rios L., Peña K., Ugás R., Quíñonez L., Mueller-Seitz E., Petz M. 2013. Compositional characterization of native Peruvian chili peppers (Capsicum spp.). J Agric Food Chem. 61(10):2530–2537.
Morales-Soriano E., Kebede B., Ugás R., Grauwet T., Van Loey A., Hendrickx M. 2018. Flavor characterization of native Peruvian chili peppers through integrated aroma fingerprinting and pungency profiling. Food Res Int. 109:250–259.
Sebastian R., Lee KY., Cho GT., Shin MJ., Kim SH., Hyun DY., Lee JR. 2019. The complete chloroplast genome sequence of a Bolivian wild chili pepper, Capsicum Eximium Hunz.(Solanaceae). Mitochondr DNA Part B. 4(1):1634–1635.
Shiragaki K., Yoko Y., Tsezuko T. 2020. Phylogenetic analysis and molecular diversity of capsicum based on rDNA-ITS region. Horticulturae. 6(4):87.
Stamatakis A. 2014. RAxML version 8: a tool for phylogenetic analysis and post-analysis of large phylogenies. Bioinformatics. 30(9):1312–1313.
Tillich M., Lehwark P., Pellizzer T., Ulbricht-Jones ES., Fischer A., Bock R., Greiner S. 2017. GeSeq—versatile and accurate annotation of organelle genomes. Nucleic Acids Res. 45(W1):W6–W11.
Tripodi P., Rabanus-Wallace MT., Barchi L., Kale S., Esposito S., Acquadro A., Schaffiteiner R., van Zonneveld M., Prohens J., Diez MJ. 2021. Global range expansion history of pepper (Capsicum spp.) revealed by over 10,000 genebank accessions. Proc Natl Acad Sci USA. 118(34):e2104315118.