



Molecular typification of bacterial isolates from the hoatzin crop (*Opisthocomus hoazin*)



INTRODUCTION

The Hoatzin is one of the world's few obligate folivorous birds inhabiting the riverine forests of northern South America. It is the smallest vertebrate to have an enlarged foregut, in which plant structural carbohydrates are converted into simple sugars through microbial fermentation.

We hypothesize that this ecosystem is likely to harbor a diverse fermentative microflora.

Our aim was to characterize bacterial cultures isolated from hoatzin crop contents by amplifying and sequencing 16S *rRNA* genes.

METHODOLOGY

Crop contents were used as inocula to Medium M10 (1). Pure cultures were stored frozen with glycerol (Figure 3).

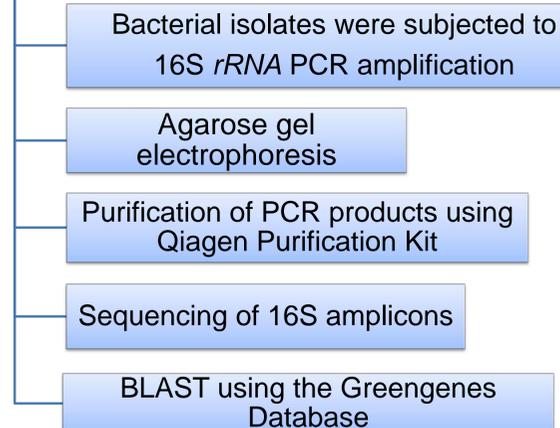


Figure 1. Map of South America and in blue the Hoatzin habitat.

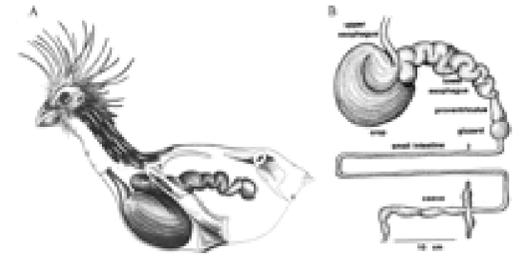


Figure 2. Schematic representation of the hoatzin digestive tract.

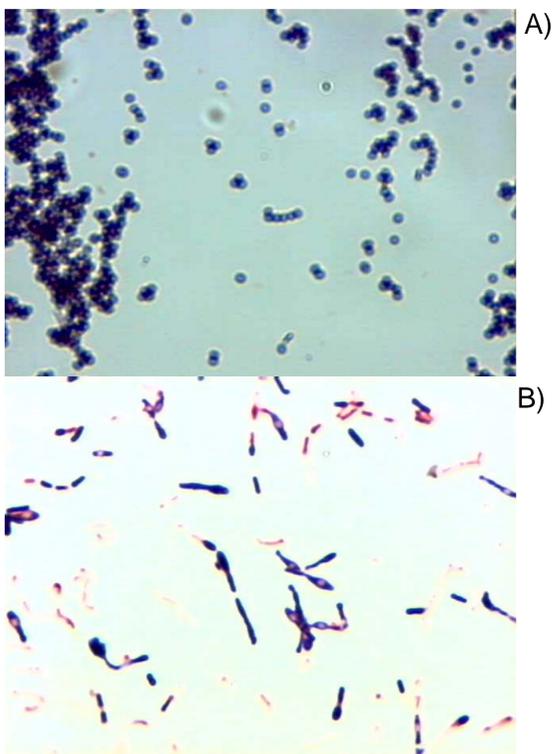


Figure 3. Pure cultures isolated from the hoatzin crop. A. Gram positive cocci B. Gram negative bacilli

RESULTS

- A total of 41 pure cultures preserved in glycerol were amplified for the 16S *rRNA* gene but only 27 were sequenced.
- The majority of the bacterial cultures were from phylum Firmicutes (Table 1) as had been previously found by cloning/sequencing (2).
- We found most bacteria to be facultative anaerobes and one specific culture is related to rumen fiber degraders (*Clostridium lituseburense*).
- Unclassified sequences (related to clones in the database) and sequences less than 97% ID to any bacteria, will be further sequenced aiming to a complete 16S gene (~1,500bp) for accurate identification.

Table 1. Bacterial diversity found in the Hoatzin crop

Phylum	Family	Genus	Species	N cultures sequenced
Proteobacteria	Enterobacteriaceae	<i>Escherichia</i>	<i>Escherichia coli</i>	2
Firmicutes	Bacillaceae	<i>Bacillus</i>	<i>Bacillus licheniformis</i>	3
	Clostridiaceae	<i>Clostridium</i>	<i>Clostridium lituseburense</i>	1
	Streptococcaceae	<i>Streptococcus</i>	<i>Streptococcus Gallolyticus</i>	4
	Enterococcaceae	<i>Enterococcus</i>	<i>Enterococcus casseliflavus</i>	1
UNCLASSIFIED	UNCLASSIFIED	UNCLASSIFIED	UNCLASSIFIED	1
UNCLASSIFIED	UNCLASSIFIED	UNCLASSIFIED	UNCLASSIFIED	15
TOTAL CULTURES				27

REFERENCES

- 1- Caldwell, D.R. and Bryant, M.P. (1966) Medium Without Rumen Fluid for Nonselective Enumeration and Isolation of Rumen Bacteria 14(5): 794-801
- 2- F. Godoy-Vitorino, R. E. Ley, Z. Gao, Z. Pei, *et al.* Appl. Envir. Microbiol.74, 19 : 5905- 5912 (2008).

CONCLUSION

The bacteria isolated from the hoatzin crop mostly resemble those found in the human gut and the cow rumen pointing to the importance of molecular culture-independent ecology studies for assessing bacterial diversity in such a unique fermentative ecosystem.

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