



SEQUENCE ANALYSIS OF 16S RDNA GENE IN POULTRY *AEROMONAS* ISOLATES

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ABSTRACT

Aeromonas species have more than one genomovar (DNA group) which cannot be separated phenotypically, so that this study was conducted to determine and differentiate genomic variation, phylogenetic relationship and closely related taxa among *Aeromonas* species on the level of 16S rRNA, there were 14 *Aeromonas* species isolated from poultry farms, 4 from poultry rations (4 *A. hydrophila*), 2 from poultry water (1 *A. hydrophila* & 1 *A. caviae*) and 8 from broiler chickens (2 *A. hydrophila* & 6 *A. caviae*). These different isolates of *Aeromonas* species from poultry farms were investigated and diagnosed by PCR then distinguished by sequence and phylogenetic tree analysis, all selected poultry *Aeromonas* isolates exhibited the specific amplification of 599 bp fragment. nucleotides sequence of 16S rRNA showed no differences in Selected Poultry *Aeromonas* isolates of the same source but differed in isolates of different sources (water, ration and broiler), *A. hydrophila* water was sub-branched and identical to *Aeromonas jandaei* strain RW102. *A. caviae* water and *A. caviae* broiler 6 were closely related to *Aeromonas caviae* strain T25, *A. caviae* broiler 1 was identical to *Aeromonas veronii* strain MS-18, *A. caviae* broiler 2,3 Were matched to *Aeromonas salmonicida* subsp. *salmonicida* 01-B526 and *Aeromonas veronii* strain 1816, *A. caviae* broiler 4, 5 were clustered with *Aeromonas veronii* strain HB18, *A. hydrophila* ration 1,2 were genetically related to *Aeromonas* sp. BEG429 while *A. hydrophila* ration 3,4 were genetically related to *Aeromonas* sp. BEG034, *A. hydrophila* broiler 1,2 were genetically related to *Aeromonas hydrophila* strain FC1361.

KEYWORDS: Aeromonas, PCR sequencing, phylogenetic tree.

INTRODUCTION

The genus *Aeromonas* was considered as glucose-fermenting, facultative anaerobic, gram-negative, rod-shaped bacteria that are positive for oxidase & catalase and resistant to vibriostatic agent OD 29.^[12]

Aeromonas species were the causative agent of multiple diseases in both warm and cold blooded animals. Also considered to be the etiology of mortality problem and lesions in some poultry farms as hemorrhagic septicemia in ducks.^[5] and cellulitis in turkeys.^[11]

On the basis of gene analysis, within that species there were more than one genomovar (DNA group) which cannot be separated phenotypically. Only 5 *Aeromonas* species were recognized 15 years ago, 3 of which existed as phenospecies (*A. hydrophila*, *A. caviae* and *A. sobria*), currently comprises 26 species: *A. allosaccharophila*, *A. aquariorum*, *A. bestiarum*, *A. bivalvium*, *A. caviae*, *A. encheleia*, *A. eucrenophila*, *A. fluvialis*, *A. hydrophila*, *A. jandaei*, *A. media*, *A. molluscorum*, *A. piscicola*, *A. popoffii*, *A. rivuli*, *A. salmonicida*, *A. sanarellii*, *A. schubertii*, *A. simiae*, *A. sobria*, *A. taiwanensis*, *A. tecta*,

A. trota, *A. veronii*, and also *Aeromonas* sp. HG11 and *Aeromonas* sp. HG13 (EntericGroup501).^[6]

Therefore, in this study the nucleotides sequence of 16S rRNA gene has been used to improve our understanding of population structure and epidemiology of *Aeromonas* species isolated from poultry farms.

MATERIAL AND METHODS

Bacterial strains

The *Aeromonas* strains analysed in this study are listed in Table 1. The strains were subcultured overnight at 30°C under aerobic conditions on nutrient agar base (Oxoid). These strains obtained from Desert Research Center.

Table 1: *Aeromonas* isolates from different origins of poultry farms.

Strain	Number	Origin (Poultry farms)
<i>A. hydrophila</i>	4	Ration
<i>A. hydrophila</i>	1	Water
<i>A. hydrophila</i>	2	Broiler chickens (liver)
<i>A. caviae</i>	1	Water
<i>A. caviae</i>	6	Broiler chickens (liver)

DNA extraction

Genomic DNA was prepared according to.^[4] from cells harvested from nutrient agar and then suspended in TE buffer (10 mM Tris/HCl, 1 mM EDTA, pH 8) using DNA extraction kit (Peq Lab).

Primers used for PCR amplification and sequencing of 16S rRNA.^[3]

Forward: 5' AGAGTTTGATCATGGCTCAG 3'

Reverse: 5' GGTTACCTTGTTACGACTT 3'

Sequencing

Sequencing was done for both directions of the PCR Products using applied biosystems by ABI 3130 genetic analyser (Applied Biosystem) and the sequencing results were checked using the NCBI BLASTn for the homology search.

RESULTS AND DISCUSSION

16s rRNA gene sequencing is one of the most powerful and precise methods for determining the interrelationships of bacteria^[13], so that in this study we determined the 16s rDNA sequences of 14 *Aeromonas* species isolated from poultry farms to clarify the intragenetic structure of the genus *Aeromonas*. The PCR product revealed the specific amplification of 599 bp fragment for all selected *Aeromonas* isolates (Figure 1).

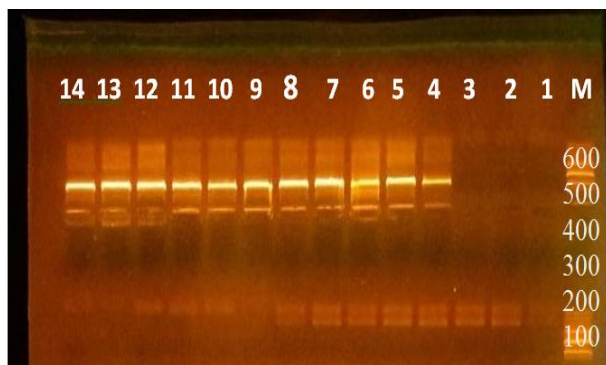


Figure.1: PCR product amplified at 599 bp.

Lane M: loader

Lane 1 - 14: *Aeromonas* species isolated from poultry farms, lane 1: *A.caviae* broiler, lane 2: *A.hydrophila* water, lane 3, 4: *A.hydrophila* ration, lane 5: *A.caviae* water, lane 6, 7: *A.hydrophila* ration, lane 8, 9: *A.hydrophila* broiler, lane 10,11,12,13,14: *A.caviae* broiler

All selected *Aeromonas* species isolated from poultry farms were genetically related when 16s rDNA sequenced, those data were in congruence with Martinez-Murcia, et al. 1992.^[10] who reported that The 13 *Aeromonas* genospecies which examined were closely related on the basis of their 16s rDNA sequences.

Nucleotides sequence of 16S rRNA showed no differences in Selected Poultry *Aeromonas* isolates of the same source but differed in isolates of different sources (water, ration and broiler) (figure. 2).

> *A. hydrophila* water

```
CCTTCGGGCCTTGCGCGATTGGATATGCCAGGTGGG
ATTAGCTAGTTGGTGAGGTAATGGCTCACCAAG
GCGACGATCCCTAGCTGGTCTGAGAGGATGATCAGCC
ACACTGGAAGTGAAGACACGGTCCAGACTCCTAC
GGGAGGCAGCAGTGGGGAATATTGCACAATGGGGGA
AACCTGATGCAGCCATGCCCGTGTGTGAAGAA
GGCCTTCGGGTTGTAAAGCACTTTCAGCGAGGAGGAA
AGGTCAGTAGCTAATATCTGCTGGCTGTGACGT
TACTCGAGAAGAAGCACCGGCTAACTCCGTGCCAGC
AGCCGCGGTAATACGGAGGGTGAAGCGTTAAT
CGGAATTACTGGGCGTAAAGCGCACGAGGCGGTTG
GATAAGTTAGATGTGAAAGCCCCGGCTCAACCT
GGGAATTGCATTTAAAAGTGTCCAGCTAGAGTCTTGT
AGAGGGGGGTAGAATTCCAGGTGTAGCGGTGAA
ATGCGTAGAGATCTGGA
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> *A. hydrophila* ration 1

```
GTGTCCTTGAGGCGTGGCTTCCGGAGCTAACGCGTTA
AATCGACCGCCTGGGGAGTACGGCCGCAAGGTT
AAAAGTCAAATGAATTGACGGGGGCCGCACAAGCG
GTGGAGCATGTGGTTTAATTCGATGCAACGCGAA
GAACCTTACCTGGCCTTGACATGTCTGGAATCCTGTA
GAGATACGGGAGTGCCTTCGGGAATCAGAACAC
AGGTGCTGCATGGCTGTCTGTCAGCTCGTGTCTGAGA
TGTTGGGTTAAGTCCCAGCAACGAGCGCAACCCC
TGTCCTTTGTTGCCAGCACGTAATGGTGGGAACTCAA
GGGAGACTGCCGGTGATAAACCAGGAGGAAGGTG
GGGATGACGTCAAGTCATCATGGCCCTTACGGCCAGG
GCTACACACGTGCTACAATGGCGCGTACAGAGG
```

> *A. hydrophila* ration

```
1GTGTCCTTGAGGCGTGGCTTCCGGAGCTAACGCGTT
AATCGACCGCCTGGGGAGTACGGCCGCAAGGTT
AAAAGTCAAATGAATTGACGGGGGCCGCACAAGCG
GTGGAGCATGTGGTTTAATTCGATGCAACGCGAA
GAACCTTACCTGGCCTTGACATGTCTGGAATCCTGTA
GAGATACGGGAGTGCCTTCGGGAATCAGAACAC
AGGTGCTGCATGGCTGTCTGTCAGCTCGTGTCTGAGA
TGTTGGGTTAAGTCCCAGCAACGAGCGCAACCCC
TGTCCTTTGTTGCCAGCACGTAATGGTGGGAACTCAA
GGGAGACTGCCGGTGATAAACCAGGAGGAAGGTG
GGGATGACGTCAAGTCATCATGGCCCTTACGGCCAGG
GCTACACACGTGCTACAATGGCGCGTACAGAGG
```

> *A. hydrophila* ration

```
1GTGTCCTTGAGGCGTGGCTTCCGGAGCTAACGCGTT
AATCGACCGCCTGGGGAGTACGGCCGCAAGGTT
AAAAGTCAAATGAATTGACGGGGGCCGCACAAGCG
GTGGAGCATGTGGTTTAATTCGATGCAACGCGAA
GAACCTTACCTGGCCTTGACATGTCTGGAATCCTGTA
GAGATACGGGAGTGCCTTCGGGAATCAGAACAC
AGGTGCTGCATGGCTGTCTGTCAGCTCGTGTCTGAGA
TGTTGGGTTAAGTCCCAGCAACGAGCGCAACCCC
TGTCCTTTGTTGCCAGCACGTAATGGTGGGAACTCAA
GGGAGACTGCCGGTGATAAACCAGGAGGAAGGTG
GGGATGACGTCAAGTCATCATGGCCCTTACGGCCAGG
GCTACACACGTGCTACAATGGCGCGTACAGAGG
```

> *A. hydrophila* ration

```
1GTGTCCTTGAGGCGTGGCTTCCGGAGCTAACGCGTT
AATCGACCGCCTGGGGAGTACGGCCGCAAGGTT
AAAAGTCAAATGAATTGACGGGGGCCGCACAAGCG
GTGGAGCATGTGGTTTAATTCGATGCAACGCGAA
GAACCTTACCTGGCCTTGACATGTCTGGAATCCTGTA
GAGATACGGGAGTGCCTTCGGGAATCAGAACAC
AGGTGCTGCATGGCTGTCTGTCAGCTCGTGTCTGAGA
TGTTGGGTTAAGTCCCAGCAACGAGCGCAACCCC
TGTCCTTTGTTGCCAGCACGTAATGGTGGGAACTCAA
GGGAGACTGCCGGTGATAAACCAGGAGGAAGGTG
```

GGGATGACGTCAAGTCATCATGGCCCTTACGGCCAGG
GCTACACACGTGCTACAATGGCGCGTACAGAGG
> *A. hydrophila* broiler 1
GGCCTTCGGGTTGTAAAGCACTTTCAGCGAGGAGGAA
AGGTCAGTAGTAATATCTGTCTGGCTGTGACGT
TACTCGCAGAAGAAGCACCCGGCTAACTCCGTGCCAGC
AGCCGCGGTAATACGGAGGGTGAAGCGTTAAT
CGGAATTACTGGGCGTAAAGCGCACGCAGGCGGTTG
GATAAGTTAGATGTGAAAGCCCCGGGCTCAACCT
GGGAATTGCATTTAAAACCTGTCCAGCTAGAGTCTTGT
AGAGGGGGGTAGAATTCCAGGTGTAGCGGTGAA
ATGCGTAGAGATCTGGAGGAATACCGGTGGCGAAGG
CGGCCCCCTGGACAAAGACTGACGCTCAGTCCGCG
AAAGCGTGGGGAGCAAACAGGATTAGATACCCTGGT
AGTCCACGCCGTAACAGATGTGCGATTGGAGGCT
GTGTCCTTGAGGCGTGGCTTCCGGAGCTAACGCGTTA
AATCGACCCGCTGGGGAGTACGGCCGAAGGTT
AAAACCTCAAATGAATTGACGGGGGCCCGCACAAGCG
GTGGAGCATGTGGTTTAATTCGATGCAACGCGAA
GAACCTTACCTGGCCTTGACATGTCTGGAATCCTGTA
GAGATACGGGAGTGCCTTCGGGAATCAGAACAC
AGGTGCTGCATGGCTGTGCTCAGCTCGTGTCTGTGAGA
TGTTGGGTTAAGTCCCAGCAACGAGCGCAACCCC
TGTCCTTTGTTGCCAGCACGTAATGGTGGGAA
> *A. hydrophila* broiler 2
GGCCTTCGGGTTGTAAAGCACTTTCAGCGAGGAGGAA
AGGTCAGTAGTAATATCTGTCTGGCTGTGACGT
TACTCGCAGAAGAAGCACCCGGCTAACTCCGTGCCAGC
AGCCGCGGTAATACGGAGGGTGAAGCGTTAAT
CGGAATTACTGGGCGTAAAGCGCACGCAGGCGGTTG
GATAAGTTAGATGTGAAAGCCCCGGGCTCAACCT
GGGAATTGCATTTAAAACCTGTCCAGCTAGAGTCTTGT
AGAGGGGGGTAGAATTCCAGGTGTAGCGGTGAA
ATGCGTAGAGATCTGGAGGAATACCGGTGGCGAAGG
CGGCCCCCTGGACAAAGACTGACGCTCAGTCCGCG
AAAGCGTGGGGAGCAAACAGGATTAGATACCCTGGT
AGTCCACGCCGTAACAGATGTGCGATTGGAGGCT
GTGTCCTTGAGGCGTGGCTTCCGGAGCTAACGCGTTA
AATCGACCCGCTGGGGAGTACGGCCGAAGGTT
AAAACCTCAAATGAATTGACGGGGGCCCGCACAAGCG
GTGGAGCATGTGGTTTAATTCGATGCAACGCGAA
GAACCTTACCTGGCCTTGACATGTCTGGAATCCTGTA
GAGATACGGGAGTGCCTTCGGGAATCAGAACAC
AGGTGCTGCATGGCTGTGCTCAGCTCGTGTCTGTGAGA
TGTTGGGTTAAGTCCCAGCAACGAGCGCAACCCC
TGTCCTTTGTTGCCAGCACGTAATGGTGGGAA
Fig. 2: Sequence analysis of 16 SrRNA for Aeromonas isolates of poultry farms.
> *A. caviae* water
AAAGCGTGGGGAGCAAACAGGATTAGATACCCTGGT
AGTCCACGCCGTAACAGATGTGCGATTGGAGGCT
GTGTCCTTGAGGCGTGGCTTCCGGAGCTAACGCGTTA
AATCGACCCGCTGGGGAGTACGGCCGAAGGTT
AAAACCTCAAATGAATTGACGGGGGCCCGCACAAGCG
GTGGAGCATGTGGTTTAATTCGATGCAACGCGAA
GAACCTTACCTGGCCTTGACATGTCTGGAATCCTGTA
GAGATACGGGAGTGCCTTCGGGAATCAGAACAC
AGGTGCTGCATGGCTGTGCTCAGCTCGTGTCTGTGAGA
TGTTGGGTTAAGTCCCAGCAACGAGCGCAACCCC
TGTCCTTTGTTGCCAGCACGTAATGGTGGGAACTCAA
GGGAGACTGCCGGTGATAAACCGGAGGAAGGTG
GGGATGACGTCAAGTCATCATGGCCCTTACGGCCAGG
GCTACACACGTGCTACAATGGCGCGTACAGAGG
GCTGCAAGCTAGCGATAGTGAGCGAATCCCAAAAAG
CGCGTCTAGTCCGGATTGGAGTCTGCAACTCGA

CTCCATGAAGTCGGAATCGCTAGTAATCGCAAATCAG
AATGTTGCGGTGAATACGTTCCCGGGCCTTGTA
> *A. caviae* broiler 1
ATTGAACGCTGGCGGCAGGCCTAACACATGCAAGTCG
AGCGGCAGCGGGAAAGTAGCTTGCTACTTTTTGC
CGCGGAGCGGCGGACGGGTGAGTAATGCCTGGGAAA
TTGCCAGTCGAGGGGGATAACAGTTGGAAACGA
CTGCTAATACCGCATAACGCCCTACGGGGAAAGCAGG
GGACCTTCGGGCCTTGC GCGATTGGATATGCC
AGGTGGGATTAGCTAGTTGGTGAGGTAATGGCTCACC
AAGGCGACGATCCCTAGCTGGTCTGAGAGGATG
ATCAGCCACACTGGAAGTGGAGACACGGTCCAGACTCC
TACGGGAGGCAGCAGTGGGAATATTGCACAAT
GGGGGAAAACCTGATGCAGCCATGCCGCGTGTGTGAA
GAAGGCCTTCGGGTTGTAAAGCACTTTCAGCGA
GGAGGAAAGGTCAGTAGCTAATATCTGCTGGCTGTGA
CGTTACTCGCAGAAGAAGCACCCGGCTAACTCCG
> *A. caviae* broiler 2
ATTGAACGCTGGCGGCAGGCCTAACACATGCAAGTCG
AGCGGCAGCGGGAAAGTAGCTTGCTACTTTTTGC
CGCGGAGCGGCGGACGGGTGAGTAATGCCTGGGAAA
TTGCCAGTCGAGGGGGATAACAGTTGGAAACGA
CTGCTAATACCGCATAACGCCCTACGGGGAAAGCAGG
GGACCTTCGGGCCTTGC GCGATTGGATATGCC
AGGTGGGATTAGCTAGTTGGTGAGGTAATGGCTCACC
AAGGCGACGATCCCTAGCTGGTCTGAGAGGATG
ATCAGCCACACTGGAAGTGGAGACACGGTCCAGACTCC
TACGGGAGGCAGCAGTGGGAATATTGCACAAT
GGGGGAAAACCTGATGCAGCCATGCCGCGTGTGTGAA
GAAGGCCTTCGGGTTGTAAAGCACTTTCAGCGA
GGAGGAAAGGTCAGTAGCTAATATCTGCTGGCTGTGA
CGTTACTCGCAGAAGAAGCACCCGGCTAACTCCG
> *A. caviae* broiler 3
ATTGAACGCTGGCGGCAGGCCTAACACATGCAAGTCG
AGCGGCAGCGGGAAAGTAGCTTGCTACTTTTTGC
CGCGGAGCGGCGGACGGGTGAGTAATGCCTGGGAAA
TTGCCAGTCGAGGGGGATAACAGTTGGAAACGA
CTGCTAATACCGCATAACGCCCTACGGGGAAAGCAGG
GGACCTTCGGGCCTTGC GCGATTGGATATGCC
AGGTGGGATTAGCTAGTTGGTGAGGTAATGGCTCACC
AAGGCGACGATCCCTAGCTGGTCTGAGAGGATG
ATCAGCCACACTGGAAGTGGAGACACGGTCCAGACTCC
TACGGGAGGCAGCAGTGGGAATATTGCACAAT
GGGGGAAAACCTGATGCAGCCATGCCGCGTGTGTGAA
GAAGGCCTTCGGGTTGTAAAGCACTTTCAGCGA
GGAGGAAAGGTCAGTAGCTAATATCTGCTGGCTGTGA
CGTTACTCGCAGAAGAAGCACCCGGCTAACTCCG
> *A. caviae* broiler 4
ATTGAACGCTGGCGGCAGGCCTAACACATGCAAGTCG
AGCGGCAGCGGGAAAGTAGCTTGCTACTTTTTGC
CGCGGAGCGGCGGACGGGTGAGTAATGCCTGGGAAA
TTGCCAGTCGAGGGGGATAACAGTTGGAAACGA
CTGCTAATACCGCATAACGCCCTACGGGGAAAGCAGG
GGACCTTCGGGCCTTGC GCGATTGGATATGCC
AGGTGGGATTAGCTAGTTGGTGAGGTAATGGCTCACC
AAGGCGACGATCCCTAGCTGGTCTGAGAGGATG
ATCAGCCACACTGGAAGTGGAGACACGGTCCAGACTCC
TACGGGAGGCAGCAGTGGGAATATTGCACAAT
GGGGGAAAACCTGATGCAGCCATGCCGCGTGTGTGAA
GAAGGCCTTCGGGTTGTAAAGCACTTTCAGCGA
GGAGGAAAGGTCAGTAGCTAATATCTGCTGGCTGTGA
CGTTACTCGCAGAAGAAGCACCCGGCTAACTCCG
> *A. caviae* broiler 5
ATTGAACGCTGGCGGCAGGCCTAACACATGCAAGTCG
AGCGGCAGCGGGAAAGTAGCTTGCTACTTTTTGC

CGGCGAGCGGCGGACGGGTGAGTAATGCCTGGGAAA
 TTGCCAGTCGAGGGGGATAACAGTTGGAAACGA
 CTGCTAATACCGCATAACGCCCTACGGGGGAAAGCAGG
 GGACCTTCGGGCCTTGC GCGATTGGATATGCC
 AGGTGGGATTAGCTAGTTGGTGAGGTAATGGCTCACC
 AAGGCGACGATCCCTAGCTGGTCTGAGAGGATG
 ATCAGCCACACTGGAAGT GAGACACGGTCCAGACTCC
 TACGGGAGGCAGCAGTGGGGAATATTGCACAAT
 GGGGGAAACCCTGATGCAGCCATGCCGCGTGTGTGAA
 GAAGGCCTTCGGGTTGTAAAGCACTTTCAGCGA
 GGAGGAAAGGTCAGTAGCTAATATCTGCTGGCTGTGA
 CGTTACTCGCAGAAGAAGCACCGGCTAACTCCG
 > A. caviae broiler 6
 ATTGAACGCTGGCGGCAGGCCTAACACATGCAAGTCG
 AGCGGCAGCGGAAAGTAGCTTGCTACTTTTTGC
 CGGCGAGCGGCGGACGGGTGAGTAATGCCTGGGAAA
 TTGCCAGTCGAGGGGGATAACAGTTGGAAACGA
 CTGCTAATACCGCATAACGCCCTACGGGGGAAAGCAGG
 GGACCTTCGGGCCTTGC GCGATTGGATATGCC
 AGGTGGGATTAGCTAGTTGGTGAGGTAATGGCTCACC
 AAGGCGACGATCCCTAGCTGGTCTGAGAGGATG
 ATCAGCCACACTGGAAGT GAGACACGGTCCAGACTCC
 TACGGGAGGCAGCAGTGGGGAATATTGCACAAT
 GGGGGAAACCCTGATGCAGCCATGCCGCGTGTGTGAA
 GAAGGCCTTCGGGTTGTAAAGCACTTTCAGCGA
 GGAGGAAAGGTCAGTAGCTAATATCTGCTGGCTGTGA
 CGTTACTCGCAGAAGAAGCACCGGCTAACTCCG

Fig. 2: Continuous.

In phylogenetic tree (figure. 3), *A. hydrophila* water was sub-branched and exhibited a closer affinity with *Aeromonas hydrophila* strain 50C1 and *Aeromonas jandaei* strain RW102, as well as *A. hydrophila* ration 1,2 were genetically clustered with *Aeromonas sp.* BEG429.

A. caviae broiler 1 was identical to *Aeromonas veronii* strain MS-18-37 and *Aeromonas caviae* GSH8M-1 in the same line with *A. hydrophila* ration 3,4 which exhibited high degree of similarity with *Aeromonas sp.* BEG034.

A. caviae broiler 2,3 were matched with *Aeromonas salmonicida* subsp. *salmonicida* 01-B526 and *Aeromonas veronii* strain 1816 in the same branch with *A. caviae* broiler 4, 5 and *A. hydrophila* broiler 1,2.

Also, on the basis of the 16s rDNA sequence analysis, *A. caviae* water and *A. caviae* broiler 6 clustered with *Aeromonas caviae* strain R25-2 and *Aeromonas caviae* strain T25-39 in a separate line matched with *Aeromonas salmonicida* subsp. *Pectinolytica*.

From the previous data all 14 *Aeromonas* isolates from poultry farms when sequenced their 16s rDNA showed closely genetic relatedness except both *A. hydrophila* and *A. caviae* of water origin and one *A. caviae* of broiler origin clustered in away separate *Aeromonas* group as well as 4 *A. caviae* broiler showed very high similarity to 2 *A. hydrophila* broiler also 4 *A. hydrophila* ration were closely related to *A. caviae* broiler 1.



Fig. 3: Phylogenetic tree based on sequencing of the 16S rRNA genes of *Aeromonas* strains.

These findings agree with Martinez-Murcia, et al. 1992.^[10] who mentioned The 16s rDNA sequences of *A. hydrophila* NCIMB 9235 exhibited no base differences with the sequence of *A. caviae* NCIMB 13016 which are in accord with previously reported high levels of chromosomal DNA relatedness between these taxa.^[7] and suggest that these organisms should be placed in a single species.

Also, Martinez-Murcia, et al. 1992.^[10] stated that *A. jandaei*, *A. schubertii* and *A. veronii* clearly represented distinct species on the basis of 16s rDNA sequence analysis results and seemed to form a separate subbranch or affiliation within the genus.

The 16s rDNA sequence of the type strain of *A. veronii* (DNA group 10) was identical to the 16s rDNA sequence of *Aeromonas sp.* strain ATCC 9071 (*A. veronii* biogroup *sobria*, DNA group 8).^[8,9] and showed that *A. veronii* biogroup *sobria* is not closely related to *A. sobria* (strains NCIMB 13016T and CIP 7433).

Six *Aeromonas* strains of animal origin (V1, V130, V183, V32, V155 and V23) as well as three strains isolated from tap water (F666I, F553E and F530D) showed identical 16S rRNA gene sequences to the *A. salmonicida* and *A. bestiarum* type strains.^[1]

The 16S rRNA gene sequence of *A. hydrophila* strain CIP 57.50 (GenBank accession no. FJ936134) was identical to those of *A. bestiarum* CIP 74.30 and *A. salmonicida* subsp. *salmonicida* NCIMB 1102 and showed a single nucleotide difference with respect to *A. salmonicida* subsp. *pectinolytica* 34 mel.^[2]

CONCLUSION

On the basis of sequencing, selected Poultry *Aeromonas* isolates of the same origin (water, ration or broiler) have the same 16S rRNA nucleotides sequence, while depending on phylogenetic tree, all 14 *Aeromonas* isolates showed closely genetic relatedness except both *A. hydrophila* and *A. caviae* of water origin and one *A. caviae* of broiler origin clustered in away separate *Aeromonas* group.

Conflict of interest

The authors declare that there is no conflict of interest regarding the publication of this article

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