Volume 10 Number 21 1982 Nucleic Acids Research

The nucleotide sequences of 5S rRNAs from a rotifer, *Brachionus plicatilis*, and two nematodes, *Rhabditis tokai* and *Caenorhabditis elegans*

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Received 4 August 1982; Revised and Accepted 24 September 1982

ABSTRACT

The nucleotide sequences of 5S rRNAs from a rotifer, *Brachionus plicatilis*, and two nematodes, *Rhabditis tokai* and *Caenorhabditis elegans* have been determined. The rotifer has two 5S rRNA species that are composed of 120 and 121 nucleotides, respectively. The sequences of these two 5S rRNAs are the same except that the latter has an additional base at its 3'-terminus. The 5S rRNAs from the two nematode species are both 119 nucleotides long. The sequence similarity percents are 79% (*Brachionus/Rhabditis*), 80% (*Brachionus/Caenorhabditis*), and 95% (*Rhabditis/Caenorhabditis*) among these three species. *Brachionus* revealed the highest similarity to *Lingula* (89%), but not to the nematodes (79%).

INTRODUCTION

In classical biology, the Rotifera and the Nematoda are considered to be phylogenically related to each other and thus often classified into one phylum, the Aschelminthes. In this paper, we have sequenced the 5S rRNAs from a rotifer, *Brachionus plicatilis*, and two nematodes, *Rhabditis tokai* and *Caenorhabditis elegans* to see if the close relationship exists in the 5S rRNA sequences between the Rotifera and the Nematoda.

MATERIALS AND METHODS

The 5S rRNAs of a rotifer, *Brachionus plicatilis*, and two nematodes, *Rhabditis tokai* [1] and *Caenorhabditis elegans* var. *Bristol*, strain N2 [2], were prepared by the phenol method from the whole organisms and purified by electrophoresis on a 15% polyacrylamide gel. The sequences were determined mainly by the chemical method of Peattie [3] using 3'-^{32}P-labelled RNA and further confirmed by the enzymatic method of Donis-Keller [4] using [3'-^{32}P] RNA. The sequences of the 5'-terminal regions of *Brachionus* and *Caenorhabditis* 5S rRNAs were confirmed by the same enzymatic method using [5'-^{32}P] RNA, and that of *Rhabditis* 5S rRNA by the formamide-fragment method of Kuchino et al. [5].

0305-1048/82/1021-7001$2.00/0

RESULTS AND DISCUSSION

Fig. 1 shows the nucleotide sequences of 5S rRNAs from *Brachionus plicatilis*, *Rhabditis tokai* and *Caenorhabditis elegans* with the sequence of *Lingula anatina* [6] for comparison. The rotifer 5S rRNA is heterogenous, one having 120 nucleotides and the other having 121 nucleotides. Their sequences are the same except that the longer one has one additional base of U at its 3'-terminus. The 5S rRNAs of the two nematodes are both 119 nucleotides long and the *Rhabditis* 5S rRNA reveals heterogeneities at 13 positions. During this study, Butler et al. [7] published the sequence of *Caenorhabditis elegans* 5S rRNA that completely agrees with our sequence determined independently.

The secondary structure of these three 5S rRNAs is essentially the same as that previously proposed for the 5S rRNAs from multicellular animals [8], where an A/C mismatch in the D-D' stem exists. The sequences of the loop regions are highly conserved (82-90% identity) while those of base-paired regions are considerably less conserved (68-74% identity) among these three species. This tendency has also been pointed out in 5S rRNAs from ciliated protozoa [9] and several eukaryotes [10].

Table 1 shows the similarity matrix of the 5S rRNA sequences from

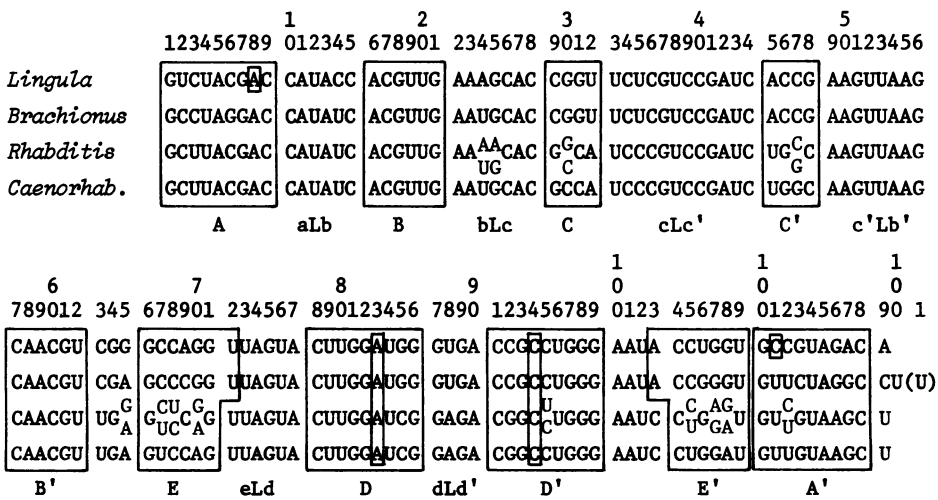

Fig. 1. Sequence alignment of 5S rRNAs of *Brachionus plicatilis*, *Rhabditis tokai*, *Caenorhabditis elegans* and *Lingula anatina* [6]. The squared-off sequences correspond to the base-paired regions in secondary structure (A,A', B,B', etc. in the lowest line). aLb, bLc, etc. are symbols for loop regions (e.g., aLb is the loop region between A and B; for these symbols, see ref. 11).

Table 1. Similarity matrix of 5S rRNA sequences of eukaryotes (%).

	VER	ART	SUR	LAN	SAN	BPL	RTO	CEL	PRO	PLA	ASC
VER*		76	81	80	80	79	73	73	64	63	60
ART*	76		83	83	77	83	71	70	64	61	59
SUR	81	83		84	84	87	76	75	66	64	63
LAN	80	83	84		87	89	76	75	66	67	65
SAN	80	77	84	87		80	68	67	65	65	64
BPL	79	83	87	89	80		78	80	63	64	63
RTO†	73	71	76	76	68	78		95	63	60	60
CEL	73	70	75	75	67	80	95		61	59	60
PRO*	64	64	66	66	65	63	63	61		61	58
PLA*	63	61	64	67	65	64	60	59	61		56
ASC*	60	59	63	65	64	63	60	60	58	56	

*The mean similarity values calculated from the sequences of 27 vertebrates (VER), 4 Arthropoda species (ART), 11 protozoa (PRO), 9 plants (PLA) and 9 Ascomycetes species (ASC). †The mean similarity values of heterogenous sequences (see Fig. 1). SUR, sea urchin; LAN, *Lingula anatina*; SAN sea anemone; BPL, *Brachionus plicatilis*; RTO, *Rhabditis tokai*; CEL, *Caenorhabditis elegans*. For the source of the sequences, see ref. 8.

eukaryotes. The 5S rRNA sequences of the two nematodes are very close despite of a considerable morphological differentiation of these two species. The rotifer is related to the nematodes only to the same extent (78-80%) as to several animals (79-83%), and is much closer to *Lingula* (89%) or sea urchin (87%). Thus the validity of classifying the Rotifera and the Nematoda into one phylum, the Aschelminthes, has not been supported by the 5S rRNA sequence data. Another point of interest is that the nematode sequences are relatively less similar to the sequences of other multicellular animal groups (70-80%) as compared with the mutual sequence similarities among the listed animal groups excluding nematodes (76-89%). Whether the Nematoda is the most anciently separated group among the listed multicellular animals must be considered when more sequence data of not only 5S rRNA but of some other informational macromolecules become available.

ACKNOWLEDGEMENTS

We thank Professor E. Nakano of Nagoya University for valuable advice and Mr. T. Kon of Fishery Center of Fukui Prefecture for providing us the rotifers used in this study. This work is supported by grants 56480377, 56570178 and 57121003 from the Ministry of Education of Japan.

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