SHORT COMMUNICATIONS

Base Composition and Genome Size of *Mycoplasma meleagridis* Deoxyribonucleic Acid

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The guanine and cytosine (GC) base compositions of deoxyribonucleic acids (DNA) of mycoplasmas have been determined by two methods: from buoyant density in CsCl by Schildkraut, Marmur & Doty (1962) and from thermal denaturation temperatures by Marmur & Doty (1962). When these methods were compared by Neimark (1967) the moles %GC of mycoplasma DNA varied among methods from as little as 26·8 to 26·5 moles %GC with *Mycoplasma mycoides* var. *mycoides* Gladysdale strain to as great as 31·7 to 35·7 moles %GC with *M. laidlawii* Sewage A strain. The base compositions of all mycoplasmas studied ranged from 23 to 24 moles %GC to as high as 39 to 40 moles %GC. Avian mycoplasmas ranged from 24·0 to 35·7 moles %GC as determined from buoyant density in CsCl by Kelton & Mandel (1969). *Mycoplasma meleagridis* strain 529 DNA had a CsCl buoyant density (g./cm.³) of 1·688 and 28·6 moles %GC. Since this was the only report on the moles %GC of *M. meleagridis* strain 529 DNA, we repeated the base composition determination from buoyant density in CsCl and compared it to results from thermal denaturation temperature.

Mycoplasma meleagridis strain 529 was supplied by R. Yamamoto, University of California, Davis, and was maintained at the FAO/WHO International Reference Centre for Animal Mycoplasma, Institute of Microbiology, University of Aarhus, Denmark. Six litres of broth medium, Brain Heart Infusion (Difco), supplemented with 1·0% fresh yeast extract, 0·8% glucose, 0·002% DNA, and 20·0% horse serum, were inoculated with 600 ml. of a 48 h. culture and incubated at 37° for 48 h. The organisms were separated at 48,200g, washed in cold saline-EDTA, lysed with sodium dodecyl sulphate, and extracted by the method of Marmur (1961).

The moles %GC calculated from buoyant density in CsCl in an analytical centrifuge and from thermal denaturation temperatures were compared. The average base composition as determined from two buoyant density experiments was 27·9 moles %GC. Buoyant densities were 1·68722 and 1·68736 g./cm.³, indicating base compositions of 27·8 and 27·9 moles %GC, respectively. Use of thermal denaturation temperatures resulted in a slightly lower average base composition of 27·0 moles %GC. Base composition was 27·0, 27·4, 27·1, and 26·3 moles %GC as determined from the temperatures corresponding to the midpoint of the 260 nm. absorbance rise; 80·36, 80·52, 80·42, and 80·10, respectively. Both methods seemed to indicate a lower base composition than the 28·1 moles %GC reported for *Mycoplasma meleagridis* strain 529 DNA. However, the maximum difference is only 1·1 moles %GC, well within the expected experimental error.

The base composition of *Mycoplasma meleagridis* places it in the lowest category (27 to 28 moles %GC) of nonfermentive mycoplasmas as created by Neimark (1967), and in the low (24·0 to 30·5 moles %GC) range of avian mycoplasma DNA as determined by Kelton & Mandel (1969).

DNA also is characterized by genome determinations. Two methods are used to study mycoplasma genome size, electron microscopic studies of the contour length of DNA by Bode & Morowitz (1967), and thermal renaturation rate by Leth Bak *et al.* (1969). Since the latter method is less tedious, it was used to determine the genome size of *Mycoplasma meleagridis* strain 529. The conditions for genome size determinations by the renaturation method were given by Wetmur & Davidson (1968).

Table 1. Values for the alkaline sedimentation coefficient, the second-order renaturation reaction rate constant (K_2) and the calculated genome size for *Mycoplasma meleagridis*

Determination	Alkaline sedimentation coefficient ($S_{20,w}^{H13}$)	K_2 value ($1 \text{ mole}^{-1} S^{-1}$)	Calculated genome size (daltons)
1	12.6	22.5	4.0×10^8
2	13.9	26.0	3.7×10^8
3	7.8	11.6	4.9×10^8
4	8.9	15.2	4.3×10^8
5	10.2	17.3	4.0×10^8
6	7.8	14.3	4.3×10^8

DNA of *Mycoplasma meleagridis* strain 529 was denatured and degraded by heating, then renatured. The rate of renaturation was recorded as the decline in absorbance at 260 nm. Six determinations of genome size of *M. meleagridis* were performed (Table 1). Mean genome size was $4.2 \pm 0.5 \times 10^8$ daltons. Although this genome size is slightly below the range, 4.4 to 4.9×10^8 daltons, of sterol-requiring *Mycoplasma* species and T-strains as determined by Leth Bak *et al.* (1969), many more experiments are required before this could be called a real difference.

Thus, *Mycoplasma meleagridis* strain 529 DNA has a GC base composition of 27.0 to 28.1 moles % and a genome size of $4.2 \pm 0.5 \times 10^8$ daltons. Both values are within the lower range of moles %GC and lower genome size exhibited by mycoplasmas.

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