Thus we observe, after the initial fast rise due to the formation of e^-_{aq}, two consecutive processes: first, a fast addition of e^-_{aq} to Fe(III)-cytochrome c, and second, the resultant slower formation of Fe(II)-cytochrome c. There may be faster intramolecular processes. With our present procedures, only the process with $k = 10^5$ sec^{-1} can be determined. The observations between 320 and 580 nm are summed up in Fig. 2.

Under the present experimental conditions the yield of e^-_{aq} is approximately six times that of H. Reactions due to H are thus of secondary importance.

From the results at 550 and 580 nm we can estimate approximately the fraction of the electrons which, having all added to the enzyme, reach the iron moiety and reduce Fe(III) to Fe(II). From the known extinction coefficients of Fe(III), Fe(II), and e^-_{aq}, the yield of e^-_{aq}, and the resultant absorbancy change, we calculate that at 550 nm 45 ± 5 percent and at 580 nm 60 ± 12 percent of the electrons produce reduction of the central Fe atom.

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DNA Complementary to Ribosomal RNA: Relation between Genomic Proportion and Ploidy

Abstract. *Ten* Nicotiana *species were assayed for the proportion of DNA that is complementary to ribosomal RNA. This proportion varies from 0.27 to 0.9 percent, with tetraploid species having lower values than the diploid species. The tetraploid species have about twice as much DNA per cell as do diploid species. Thus, the absolute number of genes for ribosomal RNA varies less than the proportion of complementary DNA. Further, the number of genes for the RNA in 80S ribosomes varies less among species than does that for the RNA in 70S ribosomes. The data indicate that loss of DNA complementary to ribosomal RNA is associated with tetraploidy in the genus* Nicotiana.

Organisms differ in the proportion of rDNA, the DNA that is complementary to ribosomal RNA (rRNA). The published values vary from 0.011 percent of total DNA for *Neoceratodus forsteri* to almost 4 percent for certain yeasts (*1, 2*), and in higher plants the proportion ranges from 0.02 percent for *Helianthus ruberosis* to 3 percent for *Cucurbita maxima* (*3–5*). A study was undertaken to determine whether this proportion might be similar for species within a genus and thus perhaps of phylogenetic or phyletic significance. Data for ten species in the genus *Nicotiana* are shown in Table 1 along with their position in the phylogenetic scheme devised for this genus by Goodspeed (*6*). Although the genomic proportion of rDNA varies considerably among the species (from 0.27 to 0.90 percent), it does not agree with the taxonomy of the genus as deduced from other characteristics. There is a substantial difference between species in all three subgenera examined and even between the species *N. paniculata* and *N. glauca* belonging to the same section. Thus, genomic proportion of rDNA appears to be a poor indicator of species relatedness, in agreement with the conclusion drawn from a similar study of the genus *Cucurbita* (*5*).

A relation between proportion of rDNA and chromosome number of the species becomes apparent, however, when the data in Table 1 are examined further. The genus *Nicotiana* contains species with either 24 or 48 chromosomes (or derivatives of these numbers), which we shall refer to, respectively, as diploid and tetraploid species. The tetraploid species examined have a genomic proportion of rDNA ranging from 0.27 to 0.43 percent, whereas the diploid species all have a higher value (0.67 to 0.90 percent). Thus, the DNA's of tetraploid species appear to have a lower proportion of rDNA than do the diploid species in this genus. *Nicotiana glauca* is a seeming exception to this rule because it has a relatively low proportion of rDNA and is reported to be a diploid species (*6*). In matter of fact this proved not to be an exception, because the plants used for DNA extraction were a local wild isolate that proved to be tetraploid when root-tip mitotic figures and flower-bud meiotic figures were examined.

The DNA's of several plant species such as pumpkin and Chinese cabbage, which have a relatively high genomic proportion of rDNA, also display a

Table 1. Genomic proportion of rDNA and chromosome number for ten *Nicotiana* species. Chromosome numbers (CN) and species arrangement are from Goodspeed (*6*). The values for rDNA were obtained by incubating an excess of ^{3}H-labeled tobacco leaf rRNA with 10 to 60 μg of alkali-denatured nuclear DNA embedded in nitrocellulose membranes; incubations were done in medium containing 0.3*M* sodium chloride and 0.03*M* sodium citrate at 68°C for 18 hours (*14*). Each value represents the average of at least two (usually more) determinations with independently prepared reagents. The listed values are within 10 percent of the most divergent determinations. Reagents were prepared as described (*7, 15*). Nuclear DNA was prepared from a 1000*g* pellet of a leaf macerate. The pellet was washed with Triton X-100 (Rohm and Haas), treated with ribonuclease and Pronase, and extracted with phenol; the resultant material was purified by preparative isopycnic banding in cesium chloride. Labeled rRNA was prepared from a macerate of leaf tissue that had been exposed to [^{3}H]uridine (*17*); the material extracted with phenol was purified by successive precipitations with ethanol and 2*M* lithium chloride (*18*). The different rRNA preparations were monitored for purity by polyacrylamide gel electrophoresis before use and had 5000 to 6000 count/min per microgram. Most populations of *N. glauca* have 24 chromosomes, but the plants used in this study had 48.

Genus *Nicotiana*	rDNA (% of total DNA)	CN
Subgenus Tabacum		
Section Genuinae		
Species *tabacum*	0.28	48
Section Tomentosae		
Species *glutinosa*	0.67	24
Subgenus Rustica		
Section Rusticae		
Species *rustica*	0.27	48
Section Paniculatae		
Species *paniculata*	0.80	24
Species *glauca*	0.32	24 (48)
Subgenus Petunioides		
Section Alatae		
Species *sylvestris*	0.90	24
Section Acuminatae		
Species *acuminata*	0.67	24
Section Bigelovianae		
Species *bigelovii*	0.32	48
Section Suaveolentes		
Species *benthamiana*	0.33	38
Species *occidentalis*	0.43	42

Table 2. Proportion of DNA complementary to the rRNA in 70*S* and 80*S* ribosomes for four *Nicotiana* species and the calculated number of cistrons per cell for each type of rRNA. The values for the proportion of DNA complementary to the rRNA in 80*S* ribosomes were obtained by hybridizing an excess of tobacco root rRNA to nuclear DNA's of the four species. This is possible because roots contain only 80*S* ribosomes (*15, 19*). Values for 70*S* rDNA's were obtained by subtracting the proportion of 80*S* rDNA from that listed in Table 1 for total proportion of rDNA (*7, 15*). The subtraction method was used for convenience and because it yields the same value for tobacco 70*S* rDNA as does the more direct determination using radioactive RNA from purified 70*S* ribosomes (*20*); CN, chromosome number.

Species	CN	DNA per nucleus (pg)	Percentage of DNA complementary to		Number of cistrons for	
			80*S* rRNA	70*S* rRNA	80*S* rRNA	70*S* rRNA
N. tabacum	48	10.1	0.15	0.13	4,500	4,600
N. occidentalis	42	9.4	0.13	0.30	3,700	10,000
N. paniculata	24	4.5	0.26	0.54	3,500	8,600
N. sylvestris	24	5.1	0.27	0.63	4,100	11,300

dense, rDNA-containing satellite component when subjected to isopycnic centrifugation in cesium chloride, whereas the DNA of *N. tabacum*, with a relatively low genomic proportion of rDNA, lacks such a visible satellite component (*4, 7*). The DNA's of the ten *Nicotiana* species examined here were subjected to analytical isopycnic centrifugation; as might have been predicted, the diploid species all displayed a clearly definable satellite component whereas none was visible for the tetraploids.

Perhaps there is a physiological requirement for a constant number of rRNA cistrons per cell, so that the different genomic proportions of rDNA may be a consequence of a constant number of cistrons but different amounts of DNA per cell. Cell DNA content was determined for diploid and tetraploid species in order to test this hypothesis. Leaf blade cells were separated with pectinase (*8*), counted with the aid of a hemocytometer, and treated to remove color and lipids (*9*); and DNA was measured (*10*). It was found, in agreement with others (*11*), that the tetraploid species *N. tabacum* contains 10.1 pg of DNA per cell and that the value for another tetraploid species, *N. occidentalis*, is 9.4 pg per cell. On the other hand, diploid species *N. sylvestris* and *N. paniculata* contain, respectively, 5.1 and 4.5 pg per cell. It is to be expected that a doubling of chromosome number leads to a doubling of cell DNA content, but the fact that the proportion of rDNA is lower in tetraploids than in diploids indicates that either rDNA does not double wth the rest of the genome during tetraploidization or that some of it is preferentially lost after the doubling event. The latter possibility seems more likely because the tetraploid species have arisen as amphidiploids (*6*).

There are two types of ribosomes in plants, chloroplastic 70*S* and cytoplasmic 80*S* ribosomes (*12*). The 70*S* ribosomes contain 23*S* and 16*S* RNA species of high molecular weight, whereas the 80*S* ribosomes contain 25*S* and 18*S* RNA species (*13*). The proportions of the genomes complementary to the RNA's of the two types of ribosomes are listed in Table 2 for four *Nicotiana* species; cell DNA content was determined along with the calculated number of cistrons per nucleus. The numbers of cistrons for the cytoplasmic rRNA's are similar for the four species, even though they have different chromosome numbers and DNA contents. However, wide variation is seen in the numbers of cistrons for the 70*S* rRNA's, the highest and lowest values differing by a factor of 2.5. The data suggest that there is a constancy in number of cistrons for cytoplasmic rRNA's in closely related plant species, even though these species may differ in chromosome number and cell DNA content; whereas the number of cistrons for the chloroplast ribosomes varies considerably.

These data, which indicate little variation in number of cistrons for 80*S* rRNA even when the genomic proportion of rDNA varies, apply to species belonging to a single genus, *Nicotiana*. However, two survey studies, one with plants (*3*) and the other with animals (*2*), demonstrate that even when species in disparate genera are compared, the genomic proportion of rDNA varies considerably more than does the actual number of rRNA cistrons.

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Aqueous Central Cavity in Aspartate Transcarbamylase from Escherichia coli

Abstract. *A three-dimensional x-ray diffraction study of aspartate transcarbamylase to 5.5-angstrom resolution, with the aid of four isomorphous heavy atom derivatives, indicates the presence of a central aqueous cavity approximating an oblate spheroid about 25 by 50 by 50 angstroms in dimension, within a molecule about 90 by 110 by 110 angstroms in largest dimensions.*

Aspartate transcarbamylase catalyzes the formation of carbamyl aspartate from carbamyl phosphate and aspartate (*1*). Cytidine triphosphate (CTP), a later product along the pathway for pyrimidine biosynthesis, inhibits this enzyme obtained from *Escherichia coli* (*2, 3*). Also, adenosine triphosphate, a