

Title	微生物の理論分類学の研究Center Speciesの新しい概念と理論的に分離された新属Halococcus
Sub Title	Studies on theoretical taxonomy of microorganisms new concept of center species and a new genus halococcus induced theoretically
Author	林, 江沢(Hayashi, Kotaku)
Publisher	共立薬科大学
Publication year	1969
Jtitle	共立薬科大学研究年報 (The annual report of the Kyoritsu College of Pharmacy). No.14 (1969.) ,p.52- 73
JaLC DOI	
Abstract	
Notes	原報
Genre	Technical Report
URL	https://koara.lib.keio.ac.jp/xoonips/modules/xoonips/detail.php?koara_id=AN00062898-00000014-0052

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微生物の理論分類学の研究
Center Species の新しい概念と理論的に分離された新属 *Halococcus*

林 江 沢

Studies on Theoretical Taxonomy of Microorganisms
New Concept of Center Species and a New Genus *Halococcus* Induced Theoretically*
Kohtaku Hayashi

I) New concept of "Center Species"

Taxonomy starts with the study of the relationship of groups to groups. Hence, the taxonomy of microorganisms should be followed in consideration with the results obtained from comparative studies of the related genera and families.

There are two concepts for the taxonomy of microorganisms that have been proposed by Sneath (1957) and by the author (1964), both of them are essentially based on Adanson's (1763) principle that all valuable features are equivalent.

I-1) Necessity of center species

The taxonomic position of a new species should be decided by comparison with the center species, and not with an optional species, of the closely related genera (Fig. 1).

As seen Fig. 1, S-value between a new isolated species '9' and species '1' is almost equal or even higher than that between species '9' and the center species '7' of group B, these relationship may lead to the conclusion that species '9' is more related to species '1' and then belongs to the group 'A'. However, comparing the group 'A' and 'B', species '9' should belong to the group (genus) 'B'.

To avoid such erroneous decision of taxonomic position of a species necessitates the introduction of a new concept in taxonomy.

I-2) Definition of center species

The center species exhibits the species having the most morphologically and biologically typical characteristics of all species in the genus to which they belong. Hence, the center species represents the characteristics of the genus.

I-3) Selection method of center species

The center species is selected by reciprocal mean similarity value (RMSV) among the species in the genus.

Formula of RMSV is shown as follows.

$$\text{RMSV} = \frac{\text{Ts}}{\text{N}-1} (\%)$$

Ts=Total sum of S-value of a species compared with another species in a genus.

N=Total number of species described above.

* The lecture summary on the Proceedings of the Tenth IAM Symposium "Taxonomy of Microorganisms" held on October, 5th, 1968, Takawana Prince Hotel, Tokyo. The Institute of Applied Microbiology. University of Tokyo.

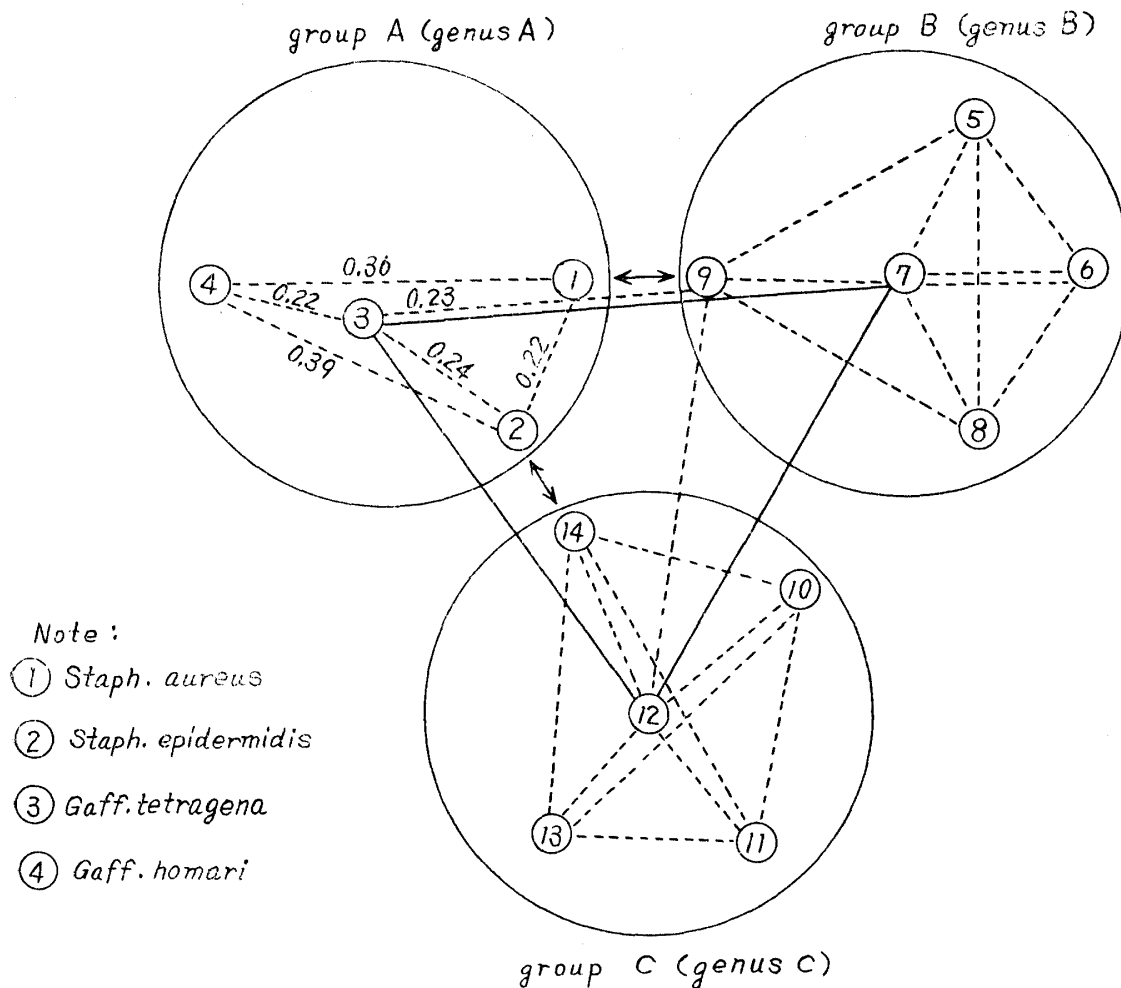


Fig. 1. The center species ③, ⑦, ⑫ and its cross-relationships of the related groups (genera) A, B, C.

Similarity value (S-value) is calculated by Sneath's method.

$$\text{Similarity value} = \frac{N_s}{N_s + N_d} (\%)$$

Center species is easily recognized because it has the highest RMSV.

I-4) Conditions to select a center species in a group (genus).

A tentative center species of a group of species is calculated from RMSV.

When all of S-value of the tentative center species against another species are 60~90%, it is a true center species and the group is considered to be genus.

If the S-value of a tentative center species for any one species is lower than 50%, it is a false center species, because the genus (group) includes heterogeneous species which should be reoriented to another genus.

These lower S-value-species should be excluded and a true center species must be selected again.

I-5) Validity of center species and RMSV

The S-value of a center species for another species in a group which may be a genus or not, and it is valuable to consider whether the group is regarded as a true genus or not.

By consideration of the S-value of the center species for another species, it is also recognized what species should be excluded (Table 2 and 4).

I-6) Taxonomic rank

The S-value and RMSV of the center species for another species (strains) or another center species are useful to consider strains, species, genus, tribe, family, order and class.

The S-value and RMSV of respective rank were shown as follows.

Species	95 ± 5 %
Genus	75 ± 15 %
Tribe	60 ± 15 %
Family	45 ± 15 %
Order	30 ± 15 %
Class	15 ± 10 %
Division	< 5 %

II Taxonomy of the family *Coccaceae*

The rearrangement of taxonomic position of species, genera, tribes and families were examined on coccoid bacteria as well as biochemically related bacillary bacteria applying the concept of author's center species with the bases of Adanson's principle and Sneath's S-value. The taxonomic characteristics described in Bergey's Manual, 7th edition were adopted as features for the calculation of S-value and RMSV.

II-1) Genera *Staphylococcus* and *Gaffkya*

Comparison of the S-value of the center species for another species of these two genera and RMSV suggested that genera *Staphylococcus* and *Gaffkya* should be integrated into one genus *Staphylococcus* (Table I).

TABLE I. Similarity value and RMSV of the genera *Staphylococcus* and *Gaffkya*

Species	1.	2.	3.	4.	RMSV (%)
1. <i>Staph. aureus</i>	100				219/3=73
2. <i>Staph. epidermidis</i>	78	100			215/3=72
3. <i>Gaff. tetragera</i>	77	76	100		231/3=77
4. <i>Gaff. homari</i>	64	61	78	100	203/3=68

Note: The bold italic species indicates the center species of the genus *Staphylococcus* which integrated genus *Gaffkya*.

II-2) Genus *Micrococcus*

As shown in table 2, this genus includes heterogeneous species. *M. flavus*, a tentative center species showed a lower S-value for some other species in the genus.

These five non-fermentative species, *M. colpogenus*, *M. cryophilus*, *M. denitrificans*, *M. morrhuae* and *M. halodenitrificans* should be excluded from the genus *Micrococcus*.

The true center species of the genus *Micrococcus* which excluded five non-fermentative species is *M. varians*.

TABLE 2. Similarity value and RMSV of the genus *Micrococcus*

Species	1.	2.	3.	4.	5.	6.	7.	8.	9.	10.	11.	12.	13.	14.	15.	16.	17.	RMSV (%)	RMSV (%)
1. <i>M. luteus</i>	100																	947/16=59	671/10=67
2. <i>M. ureae</i>	69	100																1002/16=63	687/10=69
3. <i>M. varians</i>	77	70	100															1028/16=64	723/10=72
4. <i>M. candidus</i>	74	75	70	100														978/16=61	680/10=68
5. <i>M. freudenreichii</i>	70	74	71	75	100													985/16=62	708/10=71
6. <i>M. caseolyticus</i>	66	66	71	68	82	100												989/16=62	687/10=69
7. <i>M. conglomeratus</i>	67	67	76	64	68	71	100											1020/16=64	681/10=68
8. <i>M. rubens</i>	53	54	65	50	57	61	54	100										856/16=54	579/10=58
9. <i>M. agilis</i>	63	68	74	67	62	60	69	69	100									1006/16=63	681/10=68
※10. <i>M. flavus</i>	68	71	75	74	81	74	84	53	72	100							※	1033/16=65	721/10=72
11. <i>M. roseus</i>	64	73	74	63	68	68	61	63	77	69	100							1022/16=64	680/10=68
12. <i>M. calpogenes</i>	59	53	58	55	52	59	65	45	56	58	64	100						916/16=57	292/ 5=58
13. <i>M. cryophilus</i>	42	54	43	50	49	47	50	42	51	54	50	52	100					785/16=49	253/ 5=51
14. <i>M. denitrificans</i>	47	63	53	55	50	50	55	42	53	52	61	67	50	100				890/16=56	309/ 5=62
15. <i>M. morrhuae</i>	41	52	51	48	42	50	62	54	59	53	61	62	52	61	100			892/16=56	319/ 5=64
16. <i>M. halodenitrificans</i>	44	52	50	52	50	55	51	40	51	52	58	63	49	79	71	100		871/16=54	316/ 5=63
17. <i>S. itoralis</i>	43	41	50	38	34	41	56	54	55	43	48	48	50	52	73	54	100	780/16=49	279/ 5=55

Note: ※Tentative center species. The bold italic species indicates the true center species of the genus *Micrococcus*.

II-3) Genus *Peptococcus*

The classification of the species in the genus *Peptococcus* was found to be proper, and the *Peptostreptococcus magnus* and *Peptococcus glycinophilus* showed a higher S-value to the center species of the genus *Peptococcus* rather than that of the genus *Peptostreptococcus* (Table 3 and 8).

Hence, *Peptostreptococcus magnus* should be included in the genus *Peptococcus*.

TABLE 3. Similarity value and RMSV of the genus *Peptococcus*

Species	1.	2.	3.	4.	5.	6.	7.	8.	9.	10.	11.	12.	RMSV %	RMSV %
1. <i>P. niger</i>	100												730/10=73	797/11=72
2. <i>P. asaccharolyticus</i>	79	100											723/10=72	800/11=73
3. <i>P. activus</i>	68	69	100										708/10=71	760/11=69
4. <i>P. aerogenes</i>	91	92	94	100									797/10=80	868/11=79
5. <i>P. prevotii</i>	81	77	86	87	100								789/10=79	858/11=78
6. <i>P. anaerobius</i>	83	72	61	70	81	100							729/10=73	796/11=72
7. <i>P. variabilis</i>	80	70	62	71	77	90	100						714/10=71	781/11=71
8. <i>P. saccharolyticus</i>	71	78	71	81	82	70	69	100					735/10=74	800/11=73
9. <i>P. grigoroffii</i>	63	70	85	87	88	70	68	82	100				737/10=74	795/11=72
10. <i>P. constellatus</i>	62	61	64	70	77	71	63	71	76	100			660/10=66	727/11=66
11. <i>P. glycinophilus</i>	52	55	48	54	53	61	64	60	48	45	100		540/10=54	601/11=55
12. <i>Pst. magnus</i>	67	77	52	71	69	67	67	65	58	67	61	100		721/11=66

II-4) Genera *Sarcina* and *Zymosarcina*

As shown in table 4, five out of ten species having been included in the classical genus *Sarcina*, should be excluded from this genus, because of their lower similarity value to the center species, *S. flava*.

The excluded five species are *Sarcina ventriculi*, *S. maxima*, *S. menthanica*, *S. barkeri* and *S. litoralis*.

It was proper that the residual five aerobic and packet forming species constitute the genus *Sarcina*, because of their high S-value for *S. flava*, the center species.

The two microaerophilic, fermentative and packet forming species, *S. ventriculi* and *S. maxima*, should be elevated to the genus *Zymosarcina* from the original subgenus.

The two strictly anaerobic methane-fermentative and relatively small packet forming species, *S. methanica* and *S. barkeri* should be included in the genus *Methanococcus*.

S. litoralis should be excluded from the classical genus *Sarcina*.

It may be classified in any other genus.

Various, originally heterogeneous species have been included together in the classical genus *Sarcina*, probably owing to their morphological character of packet formation.

According to the author's method each of these species will be consequently placed in its proper genus respectively, and moreover, they have the specific correlated features of the genus (Table 4).

TABLE 4. Similarity value and RMSV of the genus *Sarcina*

Specis	1.	2.	3.	4.	5.	6.	7.	8.	9.	10	RMSV (%)		
											1~10	1~9	1~5
1. <i>S. aurantiaca</i>	100										445/9=49	407/8=51	276/4=69
2. <i>S. flava</i>	77	100									473/9=53	437/8=55	293/4=73
3. <i>S. lutea</i>	70	72	100								443/9=49	405/8=51	274/4=69
4. <i>S. ureae</i>	56	65	57	100							394/9=44	356/8=45	237/4=59
5. <i>S. hansenii</i>	73	79	75	59	100						492/9=55	449/8=56	286/4=71
6. <i>S. ventriculi</i>	38	35	38	29	47	100					346/9=38	319/8=40	
7. <i>S. maxima</i>	37	32	33	32	45	77	100				339/9=38	309/8=39	77
8. <i>S. methanica</i>	26	36	30	28	33	26	24	100			314/0=35	279/8=35	
9. <i>S. barkeri</i>	30	41	30	30	38	29	29	76	100		328/9=36	303/8=38	76
10. <i>S. litoralis</i>	38	36	38	38	43	27	30	35	25	100	310/9=34		

II-5) Genus *Methanococcus*

The hitherto described genus *Methanococcus* comprise only two species *M. mazei* and *M. vannielii*.

The species *S. methanica* and *S. barkeri* in the hitherto described genus *Sarcina*, were compared with *M. mazei* and *M. vannielii*.

These two species indicated a high similarity value to the center species, *M. mazei* (Table 5).

Therefore, these two species (*S. methanica* and *S. barkeri*) should be transferred into the genus *Methanococcus*.

TABLE 5. Similarity value and RMSV of the genus *Methanococcus*

Species	1.	2.	3.	4.	RMSV
1. <i>M. mazei</i>	100				230/3=77
2. <i>M. vannielii</i>	83	100			205/3=68
3. <i>S. methanica</i>	82	69	100		228/3=76
4. <i>S. barkerii</i>	65	53	77	100	195/3=65

II-6) Genera *Leuconostoc* and *Pediococcus*

As shown in table 6, the hitherto described genera *Leuconostoc* and *Pediococcus* should

TABLE 6. S. value and RMSV of the genus *Leuconostoc* and *Pediococcus*

Species	1.	2.	3.	4.	5.	RMSV
1. <i>L. mesenteroides</i>	100					290/4=73
2. <i>L. dextranicum</i>	86	100				289/4=72
3. <i>L. citrovorum</i>	80	86	100			281/4=70
4. <i>P. cerevisiae</i>	61	54	52	100		247/4=62
5. <i>P. acidilactici</i>	63	63	63	80	100	269/4=67

be integrated into one genus, *Leuconostoc*.

The center species of the genus is *L. mesenteroides*.

II-7) Genera *Streptococcus* and *Diplococcus*

Twenty species of the genus *Streptococcus* constituted one genus.

Diplococcus pneumoniae should be included in the genus *Streptococcus*, because of its high S-value for the center species, *Streptococcus bovis* in the genus *Streptococcus* (Table 7).

II-8) Genus *Peptostreptococcus*

Peptostreptococcus magunus may be excluded from the genus *Peptostreptococcus*, and transferred to the genus *Peptococcus*, because this species showed a lower S-value to the center species of the genus *Peptostreptococcus* and a higher S-value to the center species of the genus *Peptococcus* (Table 3 and 8).

II-9) The relationship of the tribes *Streptococceae*, *Lactobacilleae* and *Propionibacteriaceae*.

II-9-a) Genus *Lactobacillus*

All species in genera *Lactobacillus*, *Catenabacterium*, *Ramibacterium* and *Cillobacterium*, except *Cillobacterium combesii*, should be included in one genus, *Lactobacillus*.

Eubacterium crispatum may be also placed in the genus *Lactobacillus* (Table 9).

II-9-b) Genus *Cillobacterium*

Cillobacterium combesii constitutes the genus *Cillobacterium*, since this species should not be included in the genus *Lactobacillus* (Table 9).

II-9-c) Genus *Eubacterium*

Only *Eubacterium lentum* constitutes the genus *Eubacterium*. The other species of this genus should be included in the genus *Butyrivacterium*, because all of them probably produce butyric acid from carbohydrates (Table 10).

II-9-d) Genus *Butyrivacterium*

Butyrivacterium rettgeri and eighteen species in the classical genus *Eubacterium* should be included in the genus *Butyrivacterium*, because they have a high S-value reciprocally (Table 10).

II-9-e) Genus *Propionibacterium*

Bergey's classification of the genus *Propionibacterium* is proper (Table 11).

II-9-f) Genus *Zymobacterium*

Only one species, *Zymob. oroticum* is properly classified in the genus *Zymobacterium* (Table 11).

II-9-g) Constitution of genera in the tribes *Streptococceae*, *Lactobacilleae* and *Propionibacteriaceae*

From a general consideration, three genera *Lactobacillus*, *Cillobacterium* and *Eubacterium* should constitute the tribe *Lactobacilleae*.

The tribe *Streptococceae* should be separated from the classical family *Lactobacillaceae*.

The tribe *Lactobacilleae* and *Propionibacteriaceae* should properly constitute the family *Lactobacillaceae*.

TABLE 7. Similarity value and RMSV of the genera *Streptococcus* and *Diphlococcus*

Species	1.	2.	3.	4.	5.	6.	7.	8.	9.	10.	11.	12.	13.	14.	15.	16.	17.	18.	19.	20.	21.	RMSV (%)		
1. <i>Str. pyogenes</i>	100																						1321/20=67	
2. <i>Str. equisimilis</i>	87	100																						1392/20=70
3. <i>Str. equi</i>	72	83	100																					1293/20=65
4. <i>Str. zooepidemicus</i>	74	77	85	100																				1355/20=68
5. <i>Str. dysgalactiae</i>	76	78	83	81	100																			1431/20=72
6. <i>Str. anginosus</i>	73	70	65	63	61	100																		1239/20=62
7. <i>Str. sanguis</i>	77	81	65	65	71	68	100																	1432/20=72
8. <i>Str. agalactiae</i>	66	73	67	72	74	64	70	100																1371/20=69
9. <i>Str. acidominimus</i>	69	72	70	68	90	63	70	73	100															1366/20=68
10. <i>Str. mitis</i>	71	72	68	68	81	75	84	74	79	100														1472/20=74
11. <i>Str. salivarius</i>	64	61	57	58	65	63	76	61	69	84	100													1333/20=67
12. <i>Str. bovis</i>	67	65	64	76	78	64	78	77	74	83	85	100												1495/20=75
13. <i>Str. equinus</i>	63	72	72	75	79	58	71	64	82	80	71	86	100											1375/20=69
14. <i>Str. thermophilus</i>	56	60	57	61	69	53	67	66	66	77	63	79	68	100										1295/20=65
15. <i>Str. uberis</i>	60	65	58	71	74	57	71	74	69	71	69	89	70	70	100									1379/20=69
16. <i>Str. faecalis</i>	59	64	52	61	58	58	70	69	57	63	61	73	64	58	75	100								1305/20=65
17. <i>Str. durans</i>	55	61	51	59	53	56	65	64	54	61	61	73	60	60	70	72	100							1249/20=63
18. <i>Str. lactis</i>	55	63	54	59	60	55	64	64	60	63	60	70	62	64	70	76	71	100						1270/20=64
19. <i>Str. cremoris</i>	52	59	51	57	56	50	61	58	55	60	56	67	61	70	63	69	65	85	100					1202/20=60
20. <i>Str. MG</i>	64	63	57	57	72	63	84	70	55	85	76	69	56	59	60	61	61	54	54	100				1301/20=65
21. <i>D. pneumoniae</i>	69	66	62	68	72	60	74	71	71	73	73	78	61	72	71	65	57	54	53	74	100			1344/20=67

TABLE 8. Similarity value and RMSV of the genus *Peptostreptococcus*

Species	1.	2.	3.	4.	5.	6.	7.	8.	9.	10.	11.	12.	13.	14.	RMSV (%)	RMSV (%)
1. <i>Pst. anaerobius</i>	100														899/12=75	843/11=77
2. <i>Pst. foetidus</i>	80	100													816/12=68	758/11=69
3. <i>Pst. lanceolatus</i>	86	72	100												837/12=70	769/11=70
4. <i>Pst. putridus</i>	75	69	67	100											869/12=72	810/11=74
5. <i>Pst. productus</i>	85	83	71	77	100										957/12=80	897/11=82
6. <i>Pst. micros</i>	67	65	71	70	75	100									842/12=70	776/11=71
7. <i>Pst. parvulus</i>	71	60	59	65	80	75	100								819/12=68	759/11=69
8. <i>Pst. intermedius</i>	82	68	71	83	88	84	72	100							926/12=77	868/11=79
9. <i>Pst. evolutus</i>	81	71	71	79	78	69	69	85	100						874/12=73	825/11=75
10. <i>Pst. paleopneumoniae</i>	72	62	67	79	79	65	67	78	70	100					853/12=71	794/11=72
11. <i>Pst. plagarumbelli</i>	79	68	71	73	92	73	82	85	85	70	100				902/12=75	840/11=76
12. <i>Pst. morbillorum</i>	65	60	63	73	89	62	59	72	67	85	62	100			815/12=68	757/11=69
13. <i>Pst. magnus</i>	56	58	68	59	60	66	60	58	49	59	62	58	100		713/12=60	
14. <i>Pep. glycinophilus</i>	52	55	59	52	47	54	52	54	50	50	53	60	68	100		

TABLE 10. S. value and RMSV of the genera *Eubacterium* and *Butyrivacterium*

Species	1.	2.	3.	4.	5.	6.	7.	8.	9.	10.	11.	12.	13.	14.	15.	16.	17.	18.	19.	20.	RMSV (%)	
	1. <i>Eub. foedians</i>	100																				1202/19=63
2. <i>Eub. niosii</i>	59	100																			1096/19=58	1057/18=59
3. <i>Eub. obstii</i>	68	72	100																		1396/19=73	1346/18=75
4. <i>Eub. rectale</i>	72	52	71	100																	1318/19=69	1262/18=70
5. <i>Eub. quartum</i>	71	70	85	71	100																1368/19=72	1316/18=73
6. <i>Eub. pseudotortuosum</i>	86	52	70	78	80	100															1287/19=68	1239/18=69
7. <i>Eub. tortuosum</i>	78	56	78	76	76	83	100														1329/19=70	1282/18=71
8. <i>Eub. quintum</i>	52	73	86	68	86	67	67	100													1361/19=72	1306/18=73
9. <i>Eub. limosum</i>	63	45	65	61	69	67	69	65	100												1149/19=60	1108/18=62
10. <i>Eub. ethylicum</i>	66	64	79	68	79	74	73	83	60	100											1311/19=69	1265/18=70
11. <i>Eub. ureolyticum</i>	69	50	82	88	74	76	76	74	71	75	100										1340/19=71	1284/18=71
12. <i>Eub. biforme</i>	58	68	77	66	64	65	79	70	63	70	67	100									1256/19=66	1211/18=67
13. <i>Eub. nitrogenes</i>	71	50	81	78	74	79	86	69	74	67	85	75	100								1321/19=70	1266/18=70
14. <i>Eub. aerofaciens</i>	60	60	78	71	71	66	82	71	63	79	74	85	75	100							1328/19=70	1274/18=71
15. <i>Eub. parvum</i>	67	50	61	80	73	79	75	62	67	63	73	78	69	69	100						1252/19=66	1198/18=67
16. <i>Eub. disciformans</i>	51	69	72	56	75	56	63	77	53	73	55	72	55	67	71	100					1142/19=60	1107/18=62
17. <i>Eub. minutum</i>	50	52	77	62	72	48	52	92	44	60	57	40	63	54	44	41	100				1118/19=59	1059/18=59
18. <i>Eub. poeciloides</i>	52	58	71	67	76	55	57	80	54	76	69	61	61	78	58	65	87	100			1245/19=66	1189/18=66
19. <i>Butyriv. rettgeri</i>	59	57	73	77	50	58	56	64	55	56	69	64	65	71	59	36	64	64	100		1164/19=61	1097/18=61
20. <i>Eub. lentum</i>	50	39	50	56	52	48	47	55	41	46	56	45	55	54	54	35	59	56	67	100	965/19=51	

TABLE 11. Similarity value and RMSV of the family *Propionibacteriaceae*

Species	1.	2.	3.	4.	5.	6.	7.	8.	9.	10.	11.	12.	13.	14.	15.	RMSV (%)
1. <i>Propion. freudenreichii</i>	100															876/14=63 700/10=70
2. <i>P. shermanii</i>	94	100														913/14=65 741/10=74
3. <i>P. rubrum</i>	67	71	100													967/14=69 809/10=81
4. <i>P. thoenii</i>	75	77	90	100												976/14=70 810/10=81
5. <i>P. zeae</i>	67	73	86	82	100											978/14=70 810/10=81
6. <i>P. technicum</i>	64	72	83	80	88	100										977/14=70 808/10=81
7. <i>P. raffinosaceum</i>	67	71	92	86	91	89	100									1012/14=72 840/10=84
8. <i>P. petersonii</i>	62	69	82	81	79	80	88	100								944/14=67 768/10=77
9. <i>P. jensenii</i>	74	79	88	88	83	85	92	81	100							990/14=71 819/10=82
10. <i>P. arabinosum</i>	62	63	72	74	76	81	80	73	74	100						918/14=66 739/10=74
11. <i>P. pentosaceum</i>	69	72	78	77	85	85	84	73	75	84	100					948/14=68 782/10=78
12. <i>Butyrib. rettgeri</i>	49	46	43	46	43	45	45	41	46	45	40	100				642/14=46
13. <i>Zymob. oroticum</i>	35	38	46	43	50	51	48	50	46	51	50	40	100			624/14=45
14. <i>Cillob. combesii</i>	36	38	32	34	32	31	33	33	33	33	35	50	34	100		509/14=36
15. <i>Eub. lentum</i>	55	50	37	43	43	43	46	52	46	50	41	63	42	35	100	646/14=46

The hitherto described family *Propionibacteriaceae* should be subordinated to the tribe *Propionibacteriaceae*.

Three genera such as *Butyribacterium*, *Propionibacterium* and *Zymobacterium* should constitute the tribe *Propionibacteriaceae*.

From a general consideration of the taxonomic system, the author suggested that the family *Lactobacillaceae* will include four tribes, namely, *Lactobacilleae*, *Propionibacterieae* and two not yet recognized tribes which shall be clarified in the future.

II-9-h) Relationship of the tribes *Streptococceae*, *Lactobacilleae* and *Propionibacterieae*

S-value and RMSV of the center species of the genera of tribes *Streptococceae*, *Lactobacilleae* and *Propionibacterieae* are shown in table 12.

TABLE 12. S. value and RMSV of the center species of the tribes *Streptococceae*, *Lactobacilleae* and *Propionibacterieae*

Center species	1.	2.	3.	4.	5.	6.	7.	8.	9.	RMSV (%)
1. <i>Leuc. mesenteroides</i>	100									370/8=46; 113/2=57
2. <i>Strep. bovis</i>	60	100			*141			*248		357/8=45; 113/2=57
3. <i>Peptostrep. productus</i>	53	53	100							407/8=41; 106/2=53
4. <i>Lact. (Ramib). pseudoramosum</i>	43	45	54	100						429/8=54; 116/2=58
5. <i>Cillob. combesii</i>	35	*131	44	58	100			*349		331/8=41; 93/2=47
6. <i>Eubact. lentum</i>	37	32	52	58	35	100				355/8=44; 93/2=47
7. <i>Propion. raffinosaceum</i>	51	54	50	58	35	45	100			393/8=49; 100/2=50
8. <i>Butyrib. (Eub). obstii</i>	33	*240	50	55	*352	50	47	100		377/8=47; 97/2=49
9. <i>Zymb. oroticum</i>	58	42	51	58	41	46	53	50	100	399/8=50; 103/2=52

Note: Mark * 1, 2, 3 are average number of nine S-value between center species of the tribe *Streptococceae* and those of the tribe *Lactobacilleae* (41%) the tribes *Streptococceae* and *Propionibacterieae* (48%), and the tribe *Lactobacilleae* and *Propionibacterieae* (49%), respectively.

II-10) Genus *Neisseria*

Bergey's classification of the genus *Neisseria* is proper and the center species is *N. flava* (Table 13).

II-11) Genus *Veillonella*

The genus *Veillonella* is also classified properly.

The center species is *V. reniformis* (Table 14).

II-12) Taxonomy of the family *Coccaceae* based on the author's concept

II-12-a) Tribe *Micrococceae*

The hitherto described family *Micrococcaceae* should be subordinated to a lower rank, to the two tribes *Micrococceae* and *Sarcineae*.

Three genera *Staphylococcus*, *Micrococcus* and *Peptococcus* constitute the tribe *Micrococceae*.

II-12-b) Tribe *Sarcineae*

The hitherto described genus *Sarcina* should be separated into two genera, namely

The relationship of the genera of these tribes are shown in the figures 2 and 3.

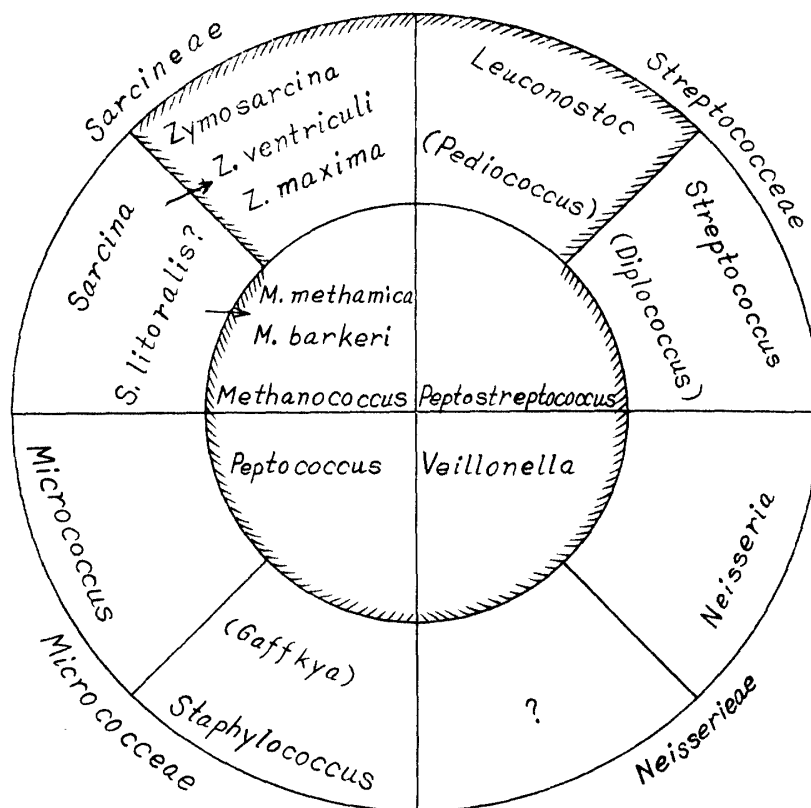


Fig. 2 Relation of the genera in the family Coccaceae

TABLE 13. Similarity value and RMSV of the genus *Neisseria*

Species	1.	2.	3.	4.	5.	6.	7.	8.	9.	10.	RMSV (%)
1. <i>N. gonorrhoeae</i>	100										463/9=51
2. <i>N. meningitidis</i>	74	100									520/9=58
3. <i>N. catarrhalis</i>	56	59	100								664/9=74
4. <i>N. flavescens</i>	50	58	89	100							652/9=72
5. <i>N. subflava</i>	52	59	82	86	100						675/9=75
6. <i>N. flava</i>	50	63	79	90	90	100					676/9=75
7. <i>N. perflava</i>	49	56	77	81	93	90	100				667/9=74
8. <i>N. sicca</i>	41	49	70	62	73	71	79	100			576/9=64
9. <i>N. haemolysans</i>	47	55	76	67	75	78	81	75	100		618/9=69
10. <i>N. caviae</i>	44	47	76	69	65	65	61	56	62	100	545/9=61

Note: The S-value of *N. subflava* to *N. perflava* is 93%.
hence these should be integrated into one species.

Sarcina and *Zymosarcina*.

The tribe *Sarcineae* comprises these two genera described above and genus *Methanococcus*.

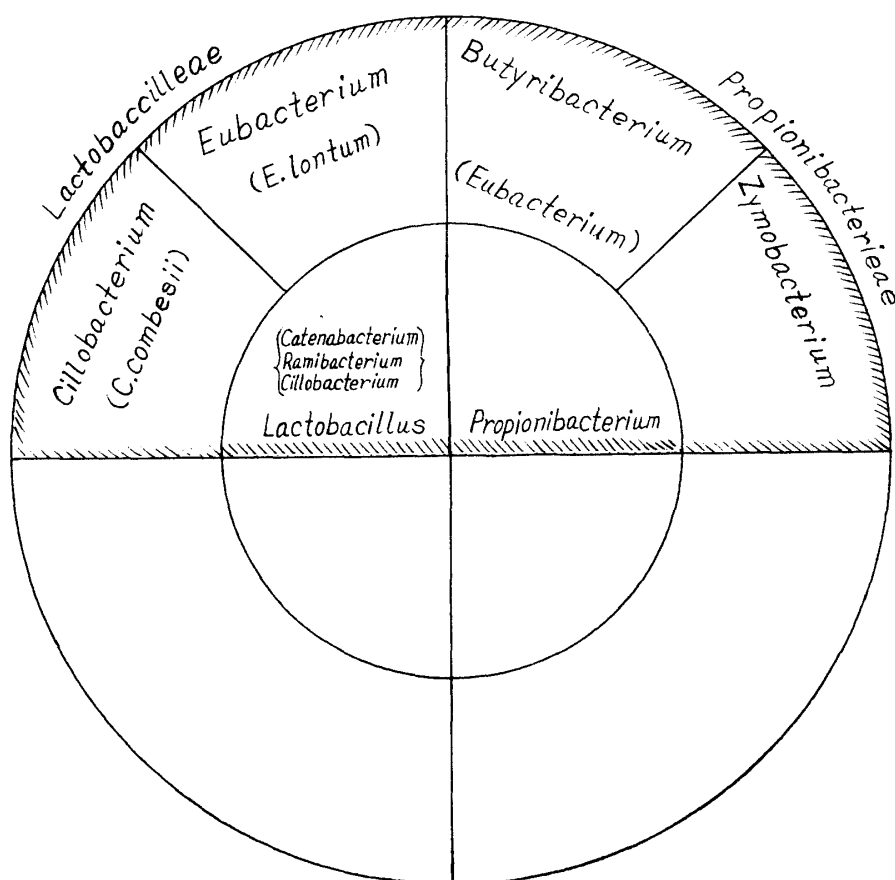


Fig. 3. Relation of the genera in the tribes Lactobacillae and Propionibacterieae

Note: The genera in the oblique lines are generally anaerobic to microaerophilic.

TABLE 14. Similarity value and RMSV of the genus *Veillonella*

Species	1.	2.	3.	4.	5.	6.	RMSV(%)
1. <i>V. parvula</i>	100						330/5=66
2. <i>V. alcalescens</i>	66	100					363/5=73
3. <i>V. discoides</i>	68	81	100				353/5=71
4. <i>V. reniformis</i>	72	73	70	100			363/5=73
5. <i>V. vulvovaginitidis</i>	52	70	66	75	100		333/5=67
6. <i>V. orbicula</i>	72	73	68	73	70	100	356/5=71

II-12-c) Tribe *Streptococceae*

The three genera, *Leuconostoc*, *Streptococcus* and *Peptostreptococcus* constitute the tribe *Streptococceae*.

II-12-d) Tribe *Neisserieae*

The classical family *Neisseriaceae* should be placed in a lower rank as tribe *Neisserieae*.

This tribe shall include three genera, but only two genera, *Neisseria* and *Veillonella* were described in literature.

Theoretically, the existence of one more genus, is speculated by the author, in the family *Coccaceae* based on the taxonomic system and relationship of this family (Fig. 2).

II-12-e) The relation of genera in the family *Coccaceae*

The comparison of the center species of genera in the family *Coccaceae* is shown in table 15.

The relation of genera and speculated genus *Halococcus* located in the tribe *Neisserieae* is shown in figures 4 and 5.

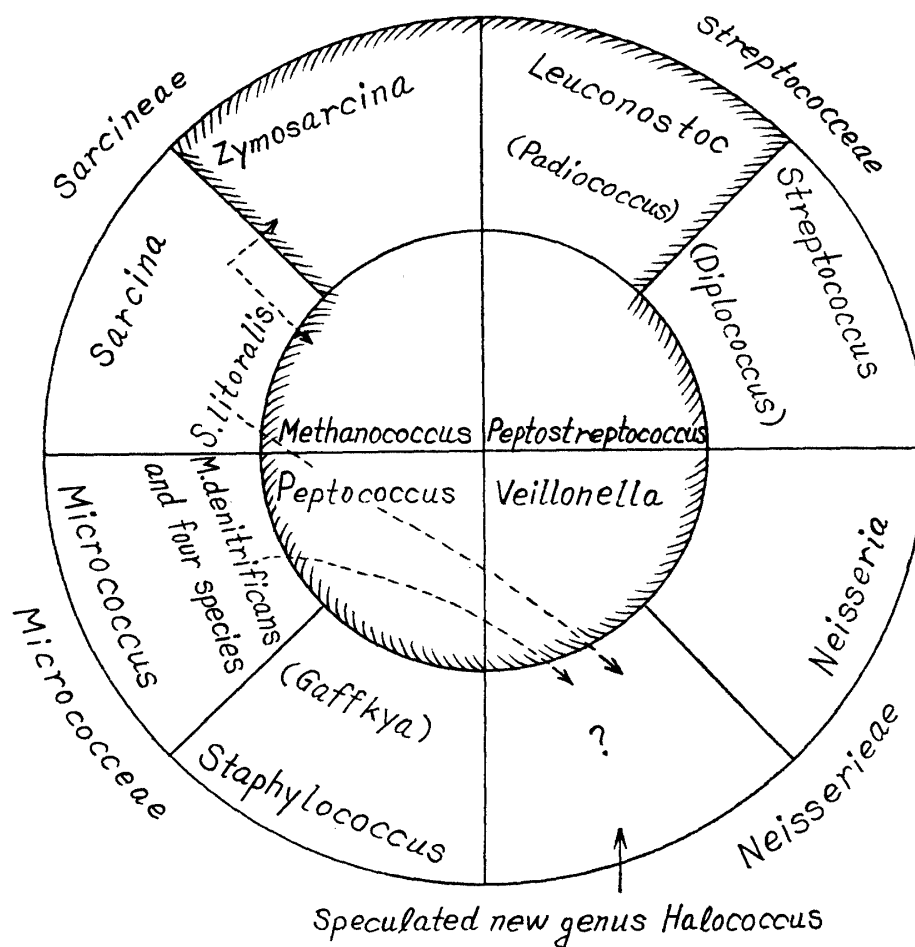


Fig. 4. Relation of the family Coccaceae and the speculated genus *Halococcus*

III A new genus *Halococcus* induced theoretically in the family *Coccaceae*

The genus *Halococcus* was induced theoretically.

This genus includes three species isolated by the author, five species having been classified in the genus *Micrococcus* and one species in the genus *Sarcina*, in literature.

TABLE 15. Comparison of the center species of genera in the family *Coccaceae*

Center species	1.	2.	3.	4.	5.	6.	7.	8.	9.	10.	11.	12.	RMSV %	
													Tribe	Family
1. <i>Staph</i> (Gaffk). <i>tetragenus</i>	100	26/44	23/39	17/43	21/45	10/33	18/38	21/39	19/40	18/36	12/25	18/39	118/2=59	528/11=48
2. <i>Microc. varians</i>	59	100	21/33	20/37	18/45	11/31	20/34	20/42	17/40	17/32	13/25	15/44	123/2=62	533/11=48
3. <i>Peptoc. aerogenes</i>	59	64	100	14/36	13/44	11/27	16/37	17/39	23/39	16/33	11/20	20/35	123/2=62	540/11=49
4. <i>Sarcina flava</i>	40	54	39	100	16/47	9/22	16/36	12/43	13/39	15/30	10/22	12/39	75/2=36	439/11=40
5. <i>Zymo. ventriculi</i>	47	40	30	34	100	10/33	21/45	20/51	18/43	14/39	8/33	11/47	64/2=32	392/11=36
6. <i>Methan. mazei</i>	30	35	41	41	30	100	8/30	8/32	10/29	9/26	7/14	9/27	71/2=36	381/11=35
7. <i>Leuc. mesenteroides</i>	47	51	43	44	47	27	100	27/47	24/46	18/33	9/31	14/40	109/2=55	487/11=44
8. <i>Sirept. bovis</i>	54	48	44	28	39	25	57	100	21/40	18/37	11/33	16/39	110/2=55	471/11=43
9. <i>Peptos. productus</i>	48	43	59	33	42	34	52	53	100	19/36	9/28	20/36	105/2=53	505/11=46
10. <i>Neisse. flava</i>	50	53	49	50	36	35	55	49	53	100	11/19	17/35	106/2=53	536/11=49
11. <i>Haloc. acetoinfaciens</i> (H•7-1)	48	52	55	45	24	50	29	33	32	57	100	10/19	110/2=55	458/11=41
12. <i>Veill. reniformis</i>	46	34	57	31	23	33	35	41	56	49	53	100	102/2=51	460/11=42

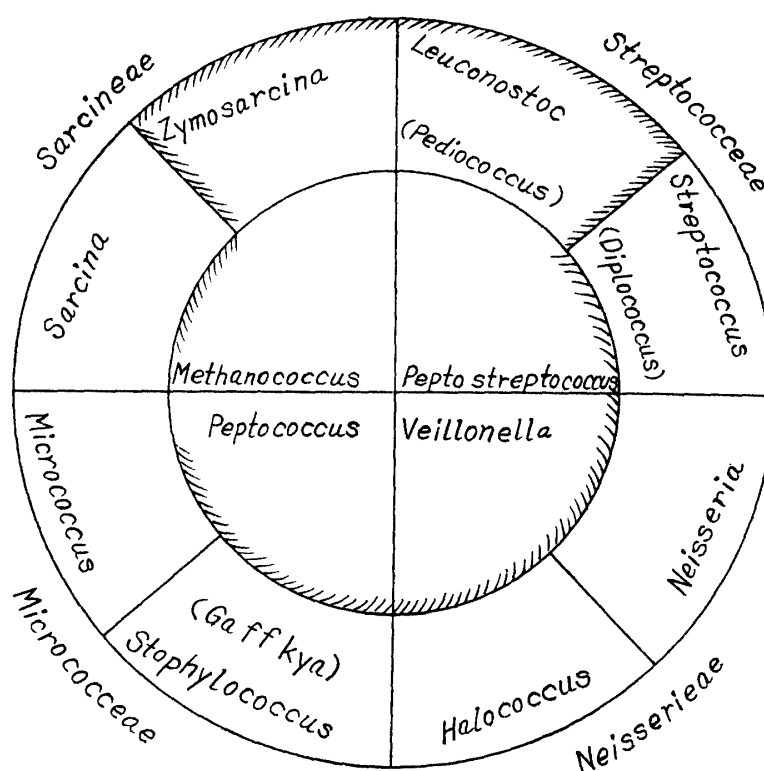


Fig. 5. Relation of the family Coccaceae

III-1) Theoretical features of the genus *Halococcus*

The theoretical features of genus *Halococcus* in the tribe *Neisserieae* were speculated as follows;

(1) Gram stain negative, (2) Spherical cells, (3) Occuring in pairs, (4) Aerobic, (5) Halotolerant, (6) Oxidase test positive, (7) No acid production from glucose.

III-2) Isolation of species from the speculated habitats.

Conscious selecting isolation was conducted using nutrient agar plate added with glucose, BTB and 5% of NaCl and checking the above mentioned features of the colonies.

Thirty-four strains were obtained from the speculated habitats such as salted fish or sea water.

III-3) Biological characteristics of species in the genus *Halococcus*

Fourteen strains were selected from thirty-four strains which had all theoretical features, and then these strains were tested for their biological characteristics.

The S-values and RMSV were calculated on the basis of one-hundred-twenty features. (Table 16).

Only three species were confirmed from the fourteen strains.

These newly recognized three species and *M. colpogenes*, *M. denitrificans* ATCC 13543, *M. morrhuae*, *M. halodenitrificans* ATCC 15174, and *Sarc. litoralis* which were excluded

TABLE 16. Similarity value of newly isolated strains and species in the new genus *Halococcus*

Strains	1. 2. 3. 4. 5. 6. 7. 8. 9. 10. 11. 12. 13. 14.														RMSV (%)			
	1. H. 7-1	100															1053/13=80	
2. H. 8-1	76	100														1022/13=79		156/2=78
3. H. 11-4	85	80	100													1186/13=91	937/10=94	165/2=83
4. H. 11-3	77	76	90	100												1134/13=87	896/10=90	
5. H. 11-2	85	80	100	90	100											1189/13=91	937/10=94	
6. H. 11-1	85	80	100	90	100	100										1189/13=91	937/10=94	
7. H. 6-2	81	80	95	90	95	95	100									1195/13=92	949/10=95	
8. H. 6-1	85	77	100	90	100	100	95	100								1198/13=92	949/10=95	
9. H. 2-2	80	74	85	88	85	85	97	97	100							1165/13=90	926/10=93	
10. H. 2-1	83	78	97	88	97	97	97	97	95	100						1193/13=82	947/10=95	
11. H. 1-3	78	76	90	90	90	90	95	90	98	93	100					1173/13=90	936/10=94	
12. H. 1-2	78	76	90	90	90	90	95	90	98	93	100	100				1173/13=90	936/10=94	
13. H. 1-1	78	76	90	90	90	90	95	90	98	93	100	100	100			1173/13=90	936/10=94	
14. H. 3-2	82	93	84	85	87	87	85	87	85	85	83	83	83	100		1109/13=85		

from the classical genera *Micrococcus* and *Sarcina*, were compared biologically.

The morphological and biological characteristics (one-hundred-twenty features) were not listed here.

The S-value and RMSV of these species were shown in table 17. These species indicate high S-value to the center species, therefore these species constitute one genus, *Halococcus*.

TABLE 17. Similarity value and RMSV of 9 species in the new genus *Halococcus*

Species	1.	2.	3.	4.	5.	6.	7.	8.	9.	RMSV (%)
1. <i>Halococcus</i> H 7-1	100←Center species									581/8=73
2. <i>Halococcus</i> H 8-1	74	100								559/8=70
3. <i>Halococcus</i> H 11-4	82	79	100							563/8=70
4. <i>Microc. colpogenes</i>	79	78	68	100						534/8=67
5. <i>Microc. denitrificans</i>	79	65	73	71	100					535/8=67
6. <i>Microc. morrhuae</i>	71	74	71	64	63	100				547/8=68
7. <i>Microc. halodenitrificans</i>	68	61	64	64	72	69	100			509/8=64
8. <i>Sarcina litoralis</i>	57	59	52	50	52	77	52	100		441/8=55
9. <i>Microc. cryophilus</i>	71	69	74	60	60	58	59	42	100	493/8=62

III-4) Congruence of theoretical features and experimental findings

In these experiments, about seventy to eighty features which were not yet reported, were tested and newly added to those described in the former reports.

It was proved that the theoretical features and the experimental findings were congruous.

Thus, it is considered that the taxonomy based on the authors concept and method mentioned above might be reasonable.

III-5) Characteristic features of the genus *Halococcus*

Gram stain negative, spherical cells, no flagella.

Aerobic, methachromatic bodies, halophilic or halotolerant, catalase positive, oxidase positive.

No acid is produced from glucose and other carbohydrates.

III-6) The differential characteristics of six species in the genus *Halococcus*

PPA (dl-phenylalanine oxidative deaminase) test, potassium nitrate reduction, urease, V.P. reaction, gelatin liquefaction, spontaneous agglutination and red pigmentation may be differential characteristics.

The differential characteristics and reaction of each species in the genus *Halococcus* are shown in table 18.

III-7) The nomenclature of three new species and emendation some species.

The new species isolated by the author were named as follows;

Halococcus strain 7-1→*Halococcus acetoinfaciens* nov. sp.

Halococcus strain 8-1→*Halococcus agglomeratus* nov. sp.

Halococcus strain 11-4→*Halococcus nondenitrificans* nov. sp.

The emended species are as follows;

TABLE 18. The differential characteristics of the six species in the genus *Halococcus*

Key features Species	PPA	KNO ₃ reduction	Urease	V.P. reaction	Gelatin liquefaction	Spontaneous agglutination	Red pigmentation
<i>Haloc. acetoinfaciens</i> (H. 7-1)	+	+	+	+	-	-	-
<i>Haloc. agglomeratus</i> (H. 8-1)	+	+	-	-	-	+	-
<i>Haloc. non-denitrificans</i> (H. 11-4)	+	-	-	-	-	-	-
<i>Haloc. denitrificans</i> (<i>M. denitrificans</i>)	-	+	-	-	-	-	-
<i>Haloc. halodenitrificans</i> (<i>M. halodenitrificans</i>)	+	+	-	-	+	-	-
<i>Haloc. cryophilus</i> (<i>M. cryophilus</i>)	+	-	+	-	-	-	-

Note: Species-names were described as newly nomenclatured or emended. The classical names were indicated in parenthesis

Micrococcus denitrificans → *Halococcus denitrificans*

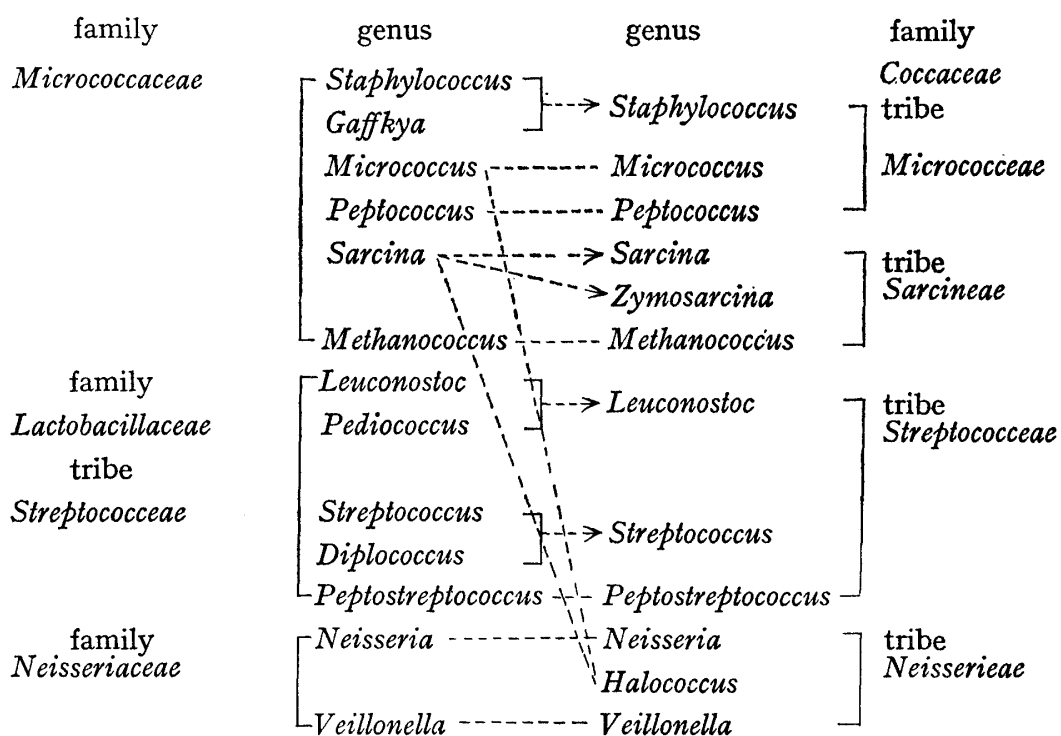
Micrococcus halodenitrificans → *Halococcus halodenitrificans*

Micrococcus cryophilus → *Halococcus cryophilus*

III-8) Relationship between the classification of Bergey's Manual and the author's proposal

Comparison between the classification of Bergey's Manual and the author's proposal is shown as follows.

The classification of Bergey's Manual The classification of the author's proposal



III-9) Key features of the family *Coccaceae*Family *Coccaceae*—spherical

- I. Tribe *Micrococceae*—Gram(+), tetrads or irregular masses, catalase(+)
1. *Staphylococcus*—facultative anaerobes, $\text{NH}_4\text{H}_2\text{PO}_4$ not utilized as a sole source of nitrogen.
 2. *Micrococcus*—aerobes, $\text{NH}_4\text{H}_2\text{PO}_4$ utilized as a sole source of nitrogen (except 2 species).
 3. *Peptococcus*—anaerobes
- II. Tribe *Sarcineae*—Gram(+), packets (except 2 species of genus *Methanococcus*)
1. *Sarcina*—aerobes, $<3 \mu$, sugar-fermentation(-), methane-production(-)
 2. *Zymosarcina*—microaerophiles or anaerobes, $>3 \mu$, sugar-fermentation(+), methane-production(-)
 3. *Methanococcus*—anaerobes, $<3 \mu$, sugar-fermentation(-), methane-production(+)
- III. Tribe *Streptococceae*—Gram(+), chains, catalase(-)
1. *Leuconostoc*—microaerophilic, l- or dl-lactic acid production(+)
 2. *Streptococcus*—facultative anaerobes, d-lactic acid production(+)
 3. *Peptostreptococcus*—anaerobes, lactic acid production(- or +)
- IV. Tribe *Neisserieae*—Gram(-), pairs
1. *Neisseria*—facultative anaerobes, sugar-fermentation(+)
 2. *Halococcus*—aerobes oxidase(+), halophilic, sugar-fermentation(-)
 3. *Veillonella*—anaerobes, sugar-fermentation (+ or -)

Conclusion

1. A new concept of center species was introduced in the taxonomy of microorganisms.
2. Theoretical taxonomy was developed on the family *Coccaceae*, based on Adanson's principle, Sneath's similarity value and the author's Concept of center species calculated from reciprocal mean similarity value.

3. A new genus *Halococcus* was induced theoretically.

It was proved that three newly recognized species and six species which had been classified in other genera, constituted the genus *Halococcus*.

4. It was interesting to found that theoretical features and experimental findings completely congruous.

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