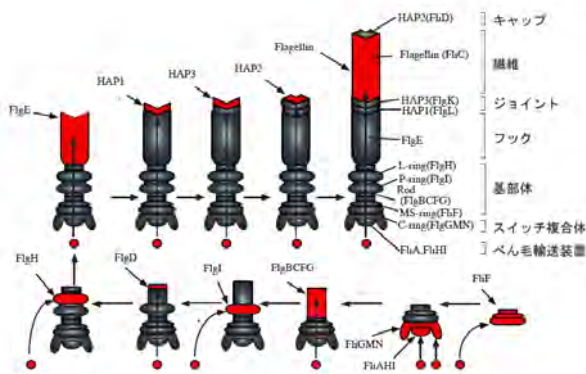
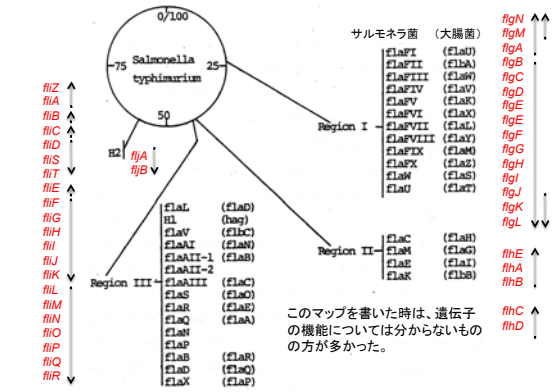


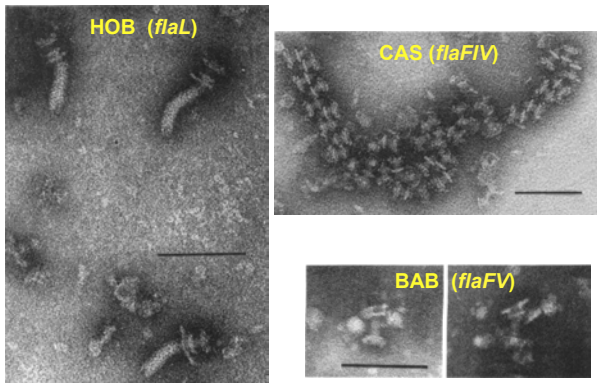
べん毛形成過程のモデル



Chromosomal map of the genes essential for flagellar formation in *Salmonella typhimurium*.

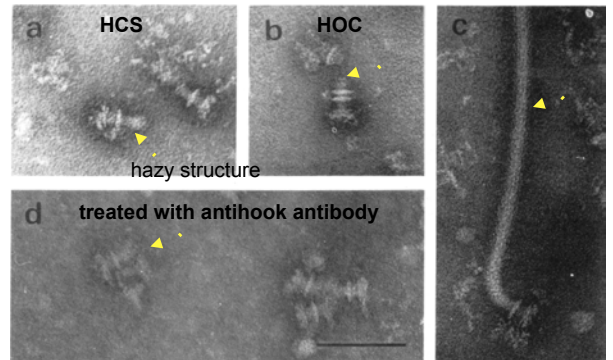


Flagellar partial structures I



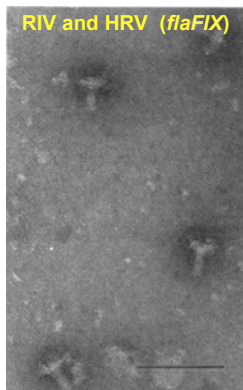
Suzuki et al., J. Bacteriol. (1978)

Flagellar partial structures II: from a *flaFVIII* mutant



Suzuki et al., J. Bacteriol. (1978)

Flagellar partial structures III



Suzuki et al., J. Bacteriol. (1978)

Frequencies of flagellar structures detected in nonflagellate mutants*

Determination	Structural entity								
	IF	HOB	BAB	CAS	RIV	RCT	HCS	HRV	HOC
Flagellate parent	+++	+	+	+ or - ^d	+	+ or -	-	-	-
<i>H1, H2</i>	-	++	+	+ or -	+	-	-	-	-
<i>flaL</i>	-	++	+	+ or -	+	-	-	-	-
<i>flaU</i>	-	++	+	+ or -	+	-	-	-	-
<i>flaR</i>	-	+++	+	+ or -	+	-	-	-	-
<i>flaFV</i>	-	-	+	+	++	++	-	-	-
<i>flaFVIII</i>	-	-	-	-	++	++	++	+ or -	+
<i>flaF1</i>	-	-	-	-	++	++	-	-	-
<i>flaFIX</i>	-	-	-	-	++	++	-	++	-
<i>flaFIV</i>	-	-	-	++	+	-	-	-	-
<i>flaF1</i>	-	-	-	-	-	-	-	-	-
<i>flaAII</i>	-	-	-	-	-	-	-	-	-
(<i>motC</i>)	++ ^f	+	+	+ or -	-	-	-	-	-
<i>flaAIII</i>	-	-	-	-	-	-	-	-	-
<i>flaB</i>	-	-	-	-	-	-	-	-	-
<i>flaC</i>	-	-	-	-	-	-	-	-	-
<i>flaD</i>	-	-	-	-	-	-	-	-	-
<i>flaE</i>	-	-	-	-	-	-	-	-	-
<i>flaFII</i>	-	-	-	-	-	-	-	-	-
<i>flaFIII</i>	-	-	-	-	-	-	-	-	-
<i>flaFVI</i>	-	-	-	-	-	-	-	-	-
<i>flaFVII</i>	-	-	-	-	-	-	-	-	-
<i>flaFX</i>	-	-	-	-	-	-	-	-	-
<i>flaK</i>	-	-	-	-	-	-	-	-	-
<i>flaM</i>	-	-	-	-	-	-	-	-	-

* The frequencies of IF and flagellar basal structures detected in fraction BMII of flagellate parents and nonflagellate mutants are shown.

^a ++. The count of each structural entity from 1/5 to 5x the count of IF in its flagellate parent.

^b +. The count of each structural entity from 1/10 to 1/5 the count of IF in its flagellate parent.

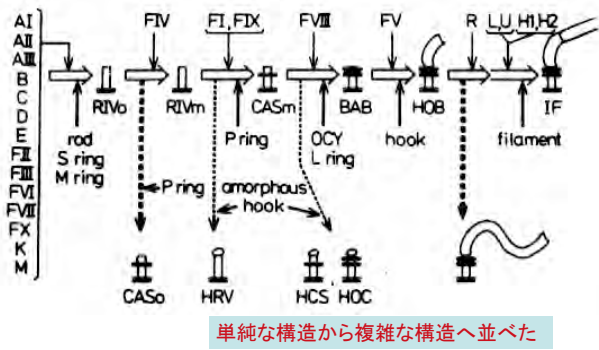
^c -. The count of each structural entity less than 1/10 the count of IF in its flagellate parent.

^d Polyhook basal body complexes were detected.

^f Paralyzed flagella, which were not discriminated morphologically from the IF of their flagellate parent.

Suzuki et al., J. Bacteriol. (1978)

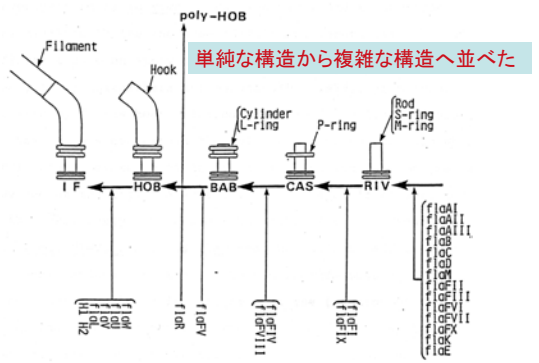
Hypothetical scheme of the pathway of flagellar morphogenesis



単純な構造から複雑な構造へ並べた

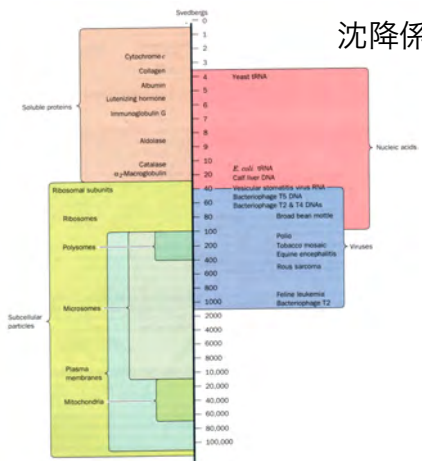
Suzuki et al., J. Bacteriol. (1978)

Stepwise process of flagellar morphogenesis in *Salmonella* inferred from the flagellar structures detected on nonflagellate mutants.

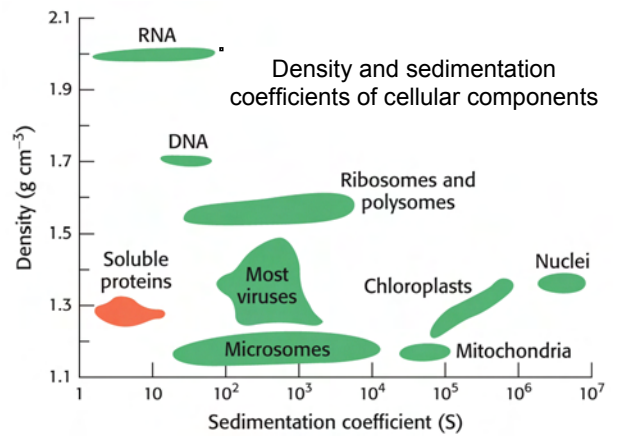


単純な構造から複雑な構造へ並べた

沈降係数



Density and sedimentation coefficients of cellular components

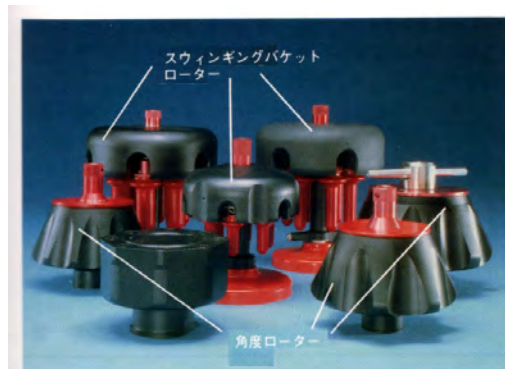


超遠心機



型式: CP100MX
 最高回転速度 (rpm): 100,000
 最大遠心加速度 (×g): 803,000
 回転制御精度 (rpm): ±10
 加速減速時間: 0~100,000rpm:5分
 温度制御精度/表示: ±0.5°C
 真空方式:
 油回転真空ポンプ+油拡散真空ポンプ
 到達圧力: 0.13Pa以下
 駆動部保証: 完全10年間
 冷却方式:
 フロンレス、サーモモジュール冷却システム
 表示:
 大きさ(mm): (W)790×(D)690×(H)1,000
 質量(Kg): 400
 標準価格(円):

超遠心ローター



50万円
 100万円
 300万円
 500万円
 800万円
 1100万円
 1500万円
 1800万円

遠心分離 I

遠心力

角速度 (rad·s⁻¹) = ω = dθ/dt

加速度 = α = ω²r 半径 = r

r = 10 cm 6,000 rpm ⇒ 0.1 · (2π · 100)² = 39,438 m/s² = 4,024 g
 30,000 rpm ⇒ 0.1 · (2π · 500)² = 985,960 m/s² = 100,608 g

沈降力は遠心力から浮力を引いたもの
 $F_s = m\omega^2 r - V\rho\omega^2 r$

Vp = 体積
 ρ = 溶液の密度
 m = 質量

摩擦力 F_r = v f
 v = 粒子の沈降速度
 f = 摩擦係数

粒子の沈降速度は沈降力と摩擦力が釣り合うまで加速する

従って $m\omega^2 r - V\rho\omega^2 r = v f$

m = M(分子量) / N(アボガドロ数)

V = 偏比容と密度の逆数

$V_p = \bar{V}m = \frac{\bar{V}M}{N}$

1gの粒子を無限大溶液媒に溶かしたときの溶液増加

20°CのDWIに蛋白質を溶かしたとき ⇒ 約0.73cm³g⁻¹

遠心分離 II

V_p = V̄ · m ; V̄ = 偏比容と密度の逆数

$V_p = \bar{V}m = \frac{\bar{V}M}{N}$. $v f = \frac{M(1 - \bar{V}\rho)\omega^2 r}{N}$

沈降係数 s を定義する

10⁻¹³s = 1S(ストベリ)として表す

$s = \frac{v}{\omega^2 r} = \frac{1}{\omega^2} \left(\frac{d \ln r}{dt} \right) = \frac{M(1 - \bar{V}\rho)}{Nf}$

加速度に対する粒子の沈降速度

半径rの粒子のf(摩擦係数)はストークの式で計算される

$f = 6\pi\eta r_p$ η = 粘度

fとfo(最小摩擦係数:水和していない球体)を求めることで分子形が推定出来る

Physical Constants of Some Proteins.

Protein	Molecular Mass (kD)	Partial Specific Volume, V _{20,w} (cm ³ · g ⁻¹)	Sedimentation Coefficient, s _{20,w} (S)	Frictional Ratio, f/f ₀
Lipase (milk)	6.7	0.714	1.14	1.190
Ribonuclease A (bovine pancreas)	12.6	0.707	2.00	1.066
Cytochrome c (bovine heart)	13.4	0.728	1.71	1.190
Myoglobin (horse heart)	16.9	0.741	2.04	1.105
α-Chymotrypsin (bovine pancreas)	21.6	0.736	2.40	1.130
Crototoxin (rattlesnake)	29.9	0.704	3.14	1.221
Concanavalin B (jack bean)	42.5	0.730	3.50	1.247
Diphtheria toxin	70.4	0.736	4.60	1.296
Cytochrome oxidase (P. aeruginosa)	89.8	0.730	5.80	1.240
Lactate dehydrogenase H (chicken)	150	0.740	7.31	1.330
Catalase (horse liver)	222	0.715	11.20	1.246
Fibrinogen (human)	340	0.725	7.63	2.336
Hemocyanin (squid)	612	0.724	19.50	1.358
Glutamate dehydrogenase (bovine liver)	1015	0.750	26.60	1.250
Turnip yellow mosaic virus protein	3013	0.740	48.80	1.470

Source: Smith, M.H., in Seber, H.A. (Ed.), Handbook of Biochemistry and Molecular Biology (2nd ed.), p. C-10, CRC Press (1970).

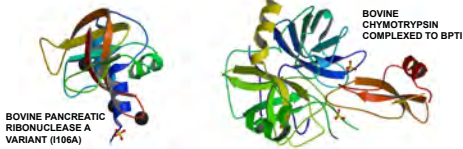
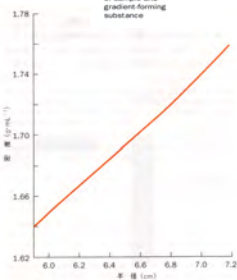
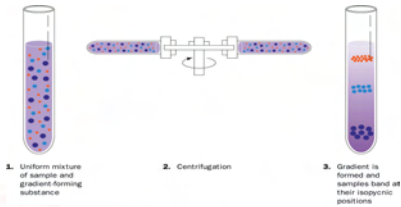


Figure 6-30 Zonal ultracentrifugation.

- (1) ゾーン超遠心分離法 (シヨ糖密度勾配)
- (2) 平衡密度勾配超遠心分離 (CsCl密度勾配)



CsCl 1.7g/mlの初期濃度で39,460 rpmで遠心する

蛋白質 = 約1.3g/ml
 DNA = 約1.7g/ml

密度勾配遠心