2013.11.13
Carl-Zeiss Lecture 2
IPHT Jena

Resonance Raman Spectroscopy; Theory and Experiment

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Landmarks of Raman Spectroscopy (HH view)

Kramers-Heisenberg-Dirac Dispersion Formula

Placzek Polarizability Theory

Albrecht Vibronic Theory

Resonance Raman Spectroscopy

Time-resolved Raman Spectroscopy

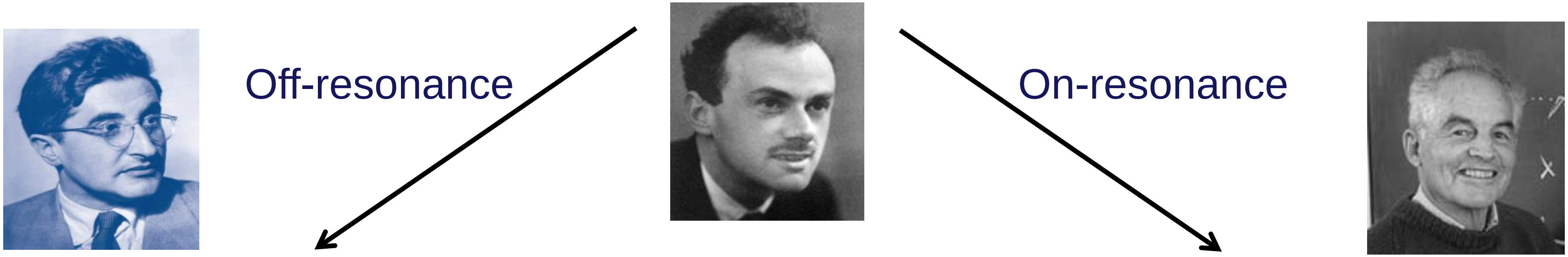
(Non-linear Raman, SERS, Raman Microspectroscopy, …. still moving)

……….

Raman microspectroscopy of Living Cells

Theoretical Framework of Raman Spectroscopy

K-H-D Dispersion Formula



Off-resonance

On-resonance

Placzek Polarizability Theory

Selection rule

$$\left(\frac{\partial \alpha_{\rho\sigma}}{\partial Q_k}\right)_0 \neq 0, \quad \Delta v = 1$$

Polarization rule

Totally symmetric mode: $0 \leq \rho < 0.75$

Non-totally symmetric mode: $\rho = 0.75$

Albrecht Vibronic Theory

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Totally symmetric mode: $\Delta v \geqq 1$

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Theory of Raman Scattering (1)

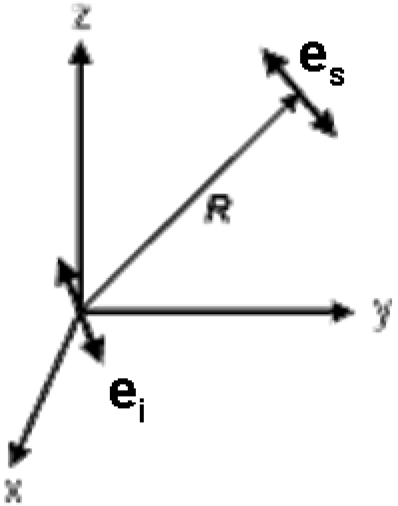
Kramers-Heisenberg-Dirac dispersion formula

P. A. M. Dirac, Principle of Quantum Mechanics


PAM Dirac
(1902-1984)

$$F_s R^2 = \frac{\omega_s^3 \omega_i}{c^4} \left| e_s \alpha e_i \right|^2 F_i$$

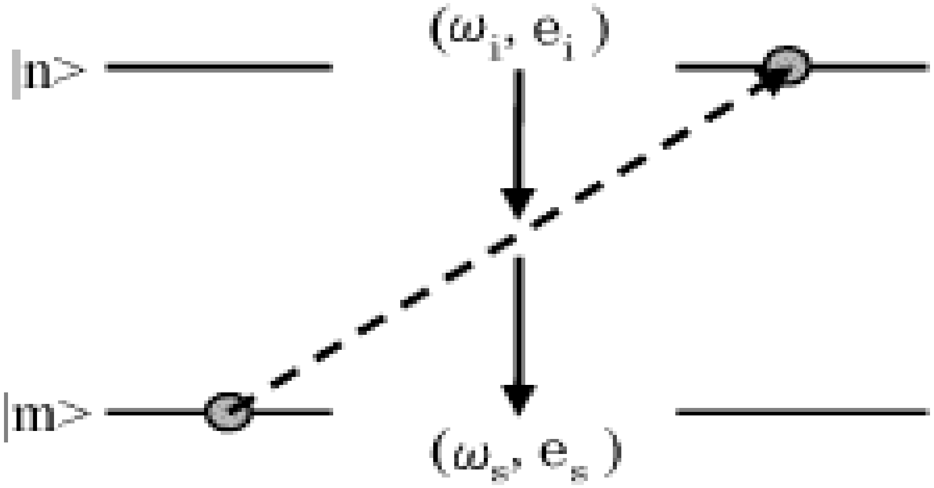
$$\alpha_{\rho\sigma} = \sum_{e \neq m,n} \left\{ \frac{<m|D_\sigma|e><e|D_\rho|n>}{E_e - E_m - E_i - i\Gamma_e} + \frac{<m|D_\rho|e><e|D_\sigma|n>}{E_e - E_n + E_i + i\Gamma_e} \right\}$$



Quantum Theory of Raman Scattering

The initial and final states of Raman scattering

In the quantum theory of Raman scattering, we calculate the probability for an optical process in which an incident photon with angular frequency ω_i and polarization vector $\boldsymbol{e}_i$ is annihilated and a new scattered photon with ω_s and $\boldsymbol{e}_s$ is created with a concomitant molecular transition from the initial state |m> to the final state |n>.



The photon number state is expressed as $|n_i,n_s>$, where n_i stands for the number of phton with ω_i and $\boldsymbol{e}_i$, ns that for ω_s and $\boldsymbol{e}_s$. The initial state |i> and the final state |f> of Raman scattering are expressed as the products of the photon and the molecular states as;

$|i>=|n_i,n_s>|m>$

$|f>=|n_i-1,n_s+1>|n>$

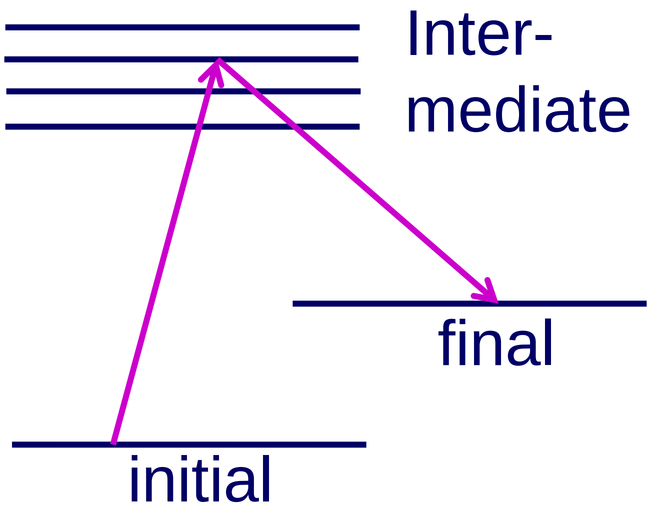
The second-order perturbation theory

$$H_0|n^{(0)}\rangle = E_n^{(0)}|n^{(0)}\rangle \quad , \quad n = 1,2,3,\cdots$$

$$H = H_0 + \lambda V$$

$$|n\rangle = |n^{(0)}\rangle + \lambda|n^{(1)}\rangle + \lambda^2|n^{(2)}\rangle + \cdots$$

$$\begin{aligned}&(H_0 + \lambda V)\left(|n^{(0)}\rangle + \lambda|n^{(1)}\rangle + \cdots\right)\\ &\quad = \left(E_n^{(0)} + \lambda E_n^{(1)} + \lambda^2 E_n^{(2)} + \cdots\right)\left(|n^{(0)}\rangle + \lambda|n^{(1)}\rangle + \cdots\right)\end{aligned}$$



Inter-
mediate

final

initial

$$|n\rangle = |n^{(0)}\rangle + \lambda\sum_{k\neq n}|k^{(0)}\rangle\frac{\langle k^{(0)}|V|n^{(0)}\rangle}{E_n^{(0)} - E_k^{(0)}} + \boxed{\lambda^2\sum_{k\neq n}\sum_{\ell\neq n}|k^{(0)}\rangle\frac{\langle k^{(0)}|V|\ell^{(0)}\rangle\langle \ell^{(0)}|V|n^{(0)}\rangle}{(E_n^{(0)} - E_k^{(0)})(E_n^{(0)} - E_\ell^{(0)})}}$$

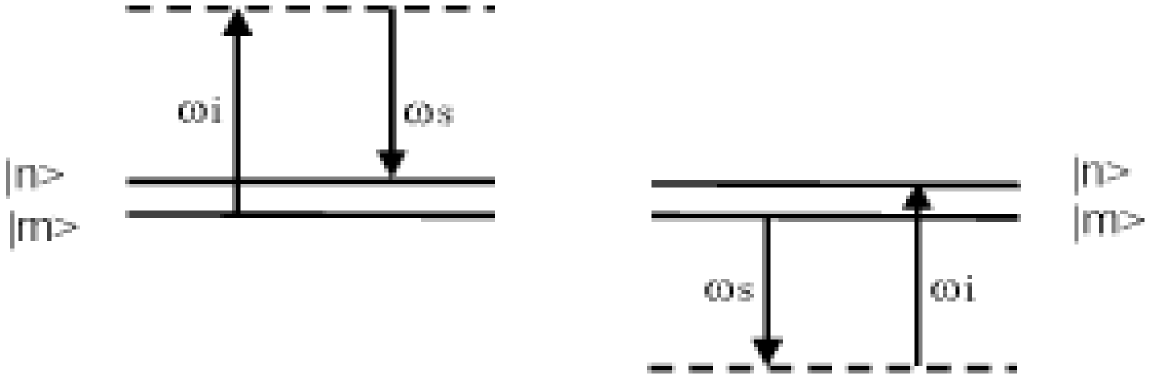
$$-\lambda^2\sum_{k\neq n}|k^{(0)}\rangle\frac{\langle n^{(0)}|V|n^{(0)}\rangle\langle k^{(0)}|V|n^{(0)}\rangle}{(E_n^{(0)} - E_k^{(0)})^2} - \frac{1}{2}\lambda^2|n^{(0)}\rangle\sum_{k\neq n}\frac{\langle n^{(0)}|V|k^{(0)}\rangle\langle k^{(0)}|V|n^{(0)}\rangle}{(E_n^{(0)} - E_k^{(0)})^2} + O(\lambda^3).$$

The intermediate states of Raman scattering

The Raman scattering process is obtained as a second order perturbation of the light-matter interaction. There are two kinds of intermediate states that can combine the initial and final states by a one-photon transition induced by a perturbation μE.

$|v_1>=|n_i-1,ns>|e>$
$|v_2>=|ni,n_s+1>|e>$

$|v_1>$ corresponds to the state in which an incident photon is annihilated with a molecular transition from |m> to |e> (figure-a, absorption resonance) and $|v_2>$ to that in which one scattered photon is created with |m> to |e> (figure-b, emission resonance).



(a) (b)

The contribution of the second intermediate state is characteristic of Raman scattering that distinguishes Raman scattering from fluorescence.

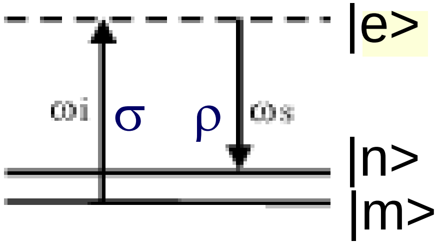
Kramers-Heisenberg-Dirac dispersion formula

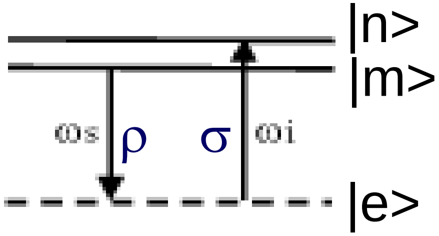
In the quantum theory of Raman scattering, it is convenient to use photon flux *F* in stead of intensity *I*, $I = h\omega C/2\pi$, where $h\omega/2\pi$ is the photon energy. *F* indicates the number of photons transmitted per unit time through unit area. The second order perturbation theory gives the following formula that connects the scattered photon number per unit time F_sR^2 and the incident photon flux F_i.

$|v_1\rangle = |n_i - 1, ns\rangle|e\rangle$ with $E_{v1} - E_m = -\hbar\omega_i + E_e - E_m = E_e - E_m - E_i$
$|v_2\rangle = |ni, n_s + 1\rangle|e\rangle$ with $E_{v2} - E_m = \hbar\omega_s + E_e - E_m = E_e - E_m - E_s = E_e - E_n + E_i$

$$F_s R^2 = \frac{\omega_s{}^3 \omega_i}{c^4} |e_s \alpha e_i|^2 F_i$$

$$\alpha_{\rho\sigma} = \sum_{e \neq m,n} \left\{ \frac{<m|D_\sigma|e><e|D_\rho|n>}{E_e - E_m - E_i - i\Gamma_e} + \frac{<m|D_\rho|e><e|D_\sigma|n>}{E_e - E_n + E_i + i\Gamma_e} \right\}$$

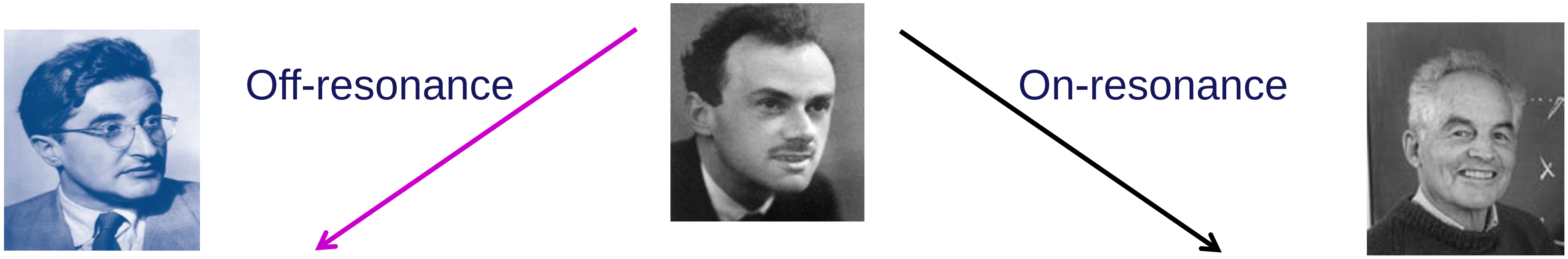




Here, e_i and e_s are the unit polarization vectors of the incident and scattered photons, $a_{\rho\sigma}$ is the Raman scattering tensor with σ and ρ being (x,y,z), and D_σ and D_ρ are the ρσ component of the electric dipole moment.

Theoretical Framework of Raman Spectroscopy

K-H-D Dispersion Formula



Placczek Polarizability Theory

Albrecht Vibronic Theory

Placzek Polarizability Theory

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Non-totally symmetric mode: $0 \leq \rho < \infty$

Placzek's polarizability theory of off-resonance Raman scattering

By introducing an adiabatic approximation, |m >=|g]|i), |n>=|g]|f), |e>=|e]|v), we obtain the formula for vibrational Raman scattering.

$$\alpha_{\rho\sigma} = (i|[g|\sum_{e\neq g}\sum_{v}\left\{\frac{[g|D_\sigma|e]|v)(v|[e|D_\rho|g]}{E_{ev}-E_{gi}-E_i-i\Gamma_e}\right.$$
$$\left.+\frac{[g|D_\rho|e]|v)(v|[e|D_\sigma|g]}{E_{ev}-E_{gf}+E_i+i\Gamma_e}\right\}|g]|f)$$

In off-resonance Raman scattering, E_{ev}-E_{gi}» E_i and therefore E_{ev}-E_{gi}-E_i is much larger than the vibrational energies. Then E_{ev}-E_{gi}-E_i+$i\Gamma$~ E_e-E_g-E_i holds with a good approximation. Then the closure property $\Sigma|v><v|=1$ simplifies the KHD formula to the following form.

$$\alpha_{\rho\sigma} \cong (i|\alpha_{\rho\sigma}|f)$$

$$\alpha_{\rho\sigma} = \sum_{e\neq g}\left\{\frac{[g|D_\sigma|e][e|D_\rho|g]}{E_e-E_g-E_i}+\frac{[g|D_\rho|e][e|D_\sigma|g]}{E_e-E_g+E_i}\right\}$$

The Raman scattering tensor component $a_{\rho\sigma}$ is approximately given by the vibrational matrix element of the polarizability tensor component $\alpha_{\rho\sigma}$ (Placzek polarizability theory).

Selection rule for off-resonance Raman scattering

The initial vibrational state |i) and the final vibrational state |f) are expressed as the products of vibratioal states,

$|i) = \Pi|v_{ki}>$ (12), $|f) = \Pi|v_{kf}>$, (13)

where v_{ki} and v_{kf} are the vibrational quantum numbers of the k-th vibrational mode in the initial and final states.

The polarizability component $\alpha_{\rho\sigma}$ is expanded into a power series of normal coordinates Q_k.

$$\alpha_{\rho\sigma} = (\alpha_0)_{\rho\sigma} + \sum_k \left(\frac{\partial \alpha_{\rho\sigma}}{\partial Q_k}\right)_0 Q_k \qquad (14)$$

Under a harmonic approximation, the vibrational matrix element of the polarizabilty component is given in the following form.

$$(i\,|\,Q_k\,|\,f) \propto \delta_{v_{ki},\,v_{kf}\pm 1} \prod_{l\neq k} \delta_{v_{li},\,v_{lf}} \qquad (15)$$

We finally obtain the selection rule of off-resonance Raman scattering.

$$\left(\frac{\partial \alpha_{\rho\sigma}}{\partial Q_k}\right)_0 \neq 0 \qquad (16) \qquad \text{and} \qquad \Delta v_k = v_{kf} - v_{ki} = \pm 1 \qquad (17)$$

Theory of Raman Scattering (2)

Polarizability Theory of Vibrational Raman Scattering

1) Off-resonance condition, 2) Non-degenerate condition

G. Placzek (1905-1955)

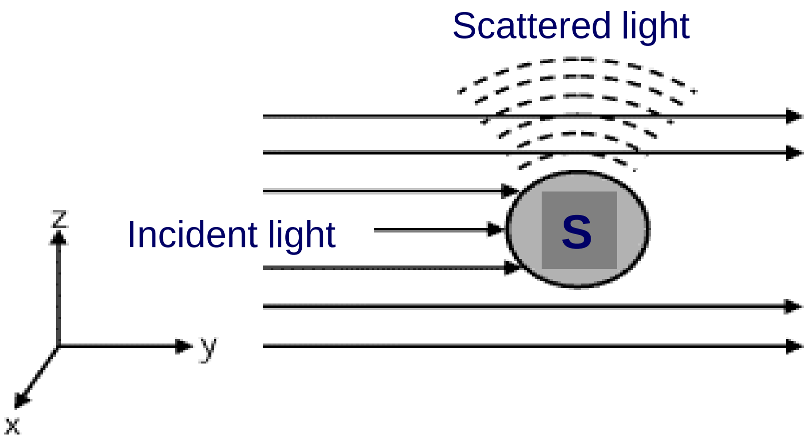
$$a_{\rho\sigma} \cong (i|\alpha_{\rho\sigma}|f)$$

$$\alpha_{\rho\sigma} = \sum_{e\neq g}\left\{\frac{[g|D_\sigma|e][e|D_\rho|g]}{E_e - E_g - E_i} + \frac{[g|D_\rho|e][e|D_\sigma|g]}{E_e - E_g + E_i}\right\}$$

Selection rule

$$\left(\frac{\partial\alpha_{\rho\sigma}}{\partial Q_k}\right)_0 \neq 0, \quad \Delta v = \pm 1$$

Depolarization ratio $\rho = I_\perp / I_{//} = I_y / I_x = \frac{3G_s+5G_a}{10G_0+4G_s}$



$a_{\rho\sigma} = a^0_{\rho\sigma} + a^a_{\rho\sigma} + a^s_{\rho\sigma}$

$G_0=\Sigma(a^0_{\rho\sigma})^2$, $G_a=\Sigma(a^a_{\rho\sigma})^2$, $G_s=\Sigma(a^s_{\rho\sigma})^2$

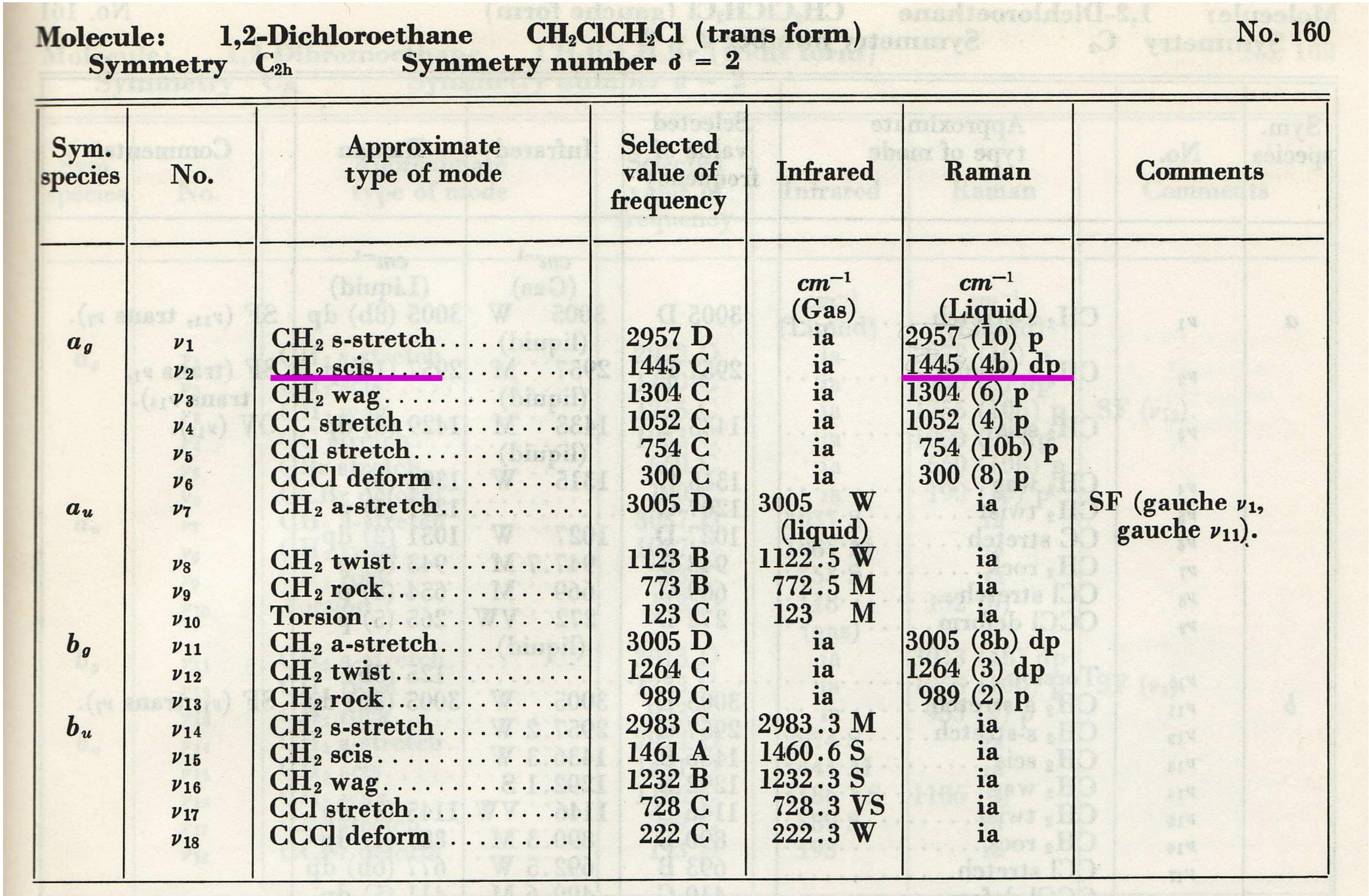
Totally symmetric mode:

$G_0\neq 0$, $G_a=0$, $G_s\neq 0$ $0\leq\rho<0.75$

Non-totally symmetric mode:

$G_0=0$, $G_a=0$, $G_s\neq 0$ $\rho=0.75$

Depolarized Totally-symmetric Raman Band?



Molecule: 1,2-Dichloroethane CH_2ClCH_2Cl (trans form) No. 160
Symmetry C_{2h} Symmetry number $\sigma = 2$

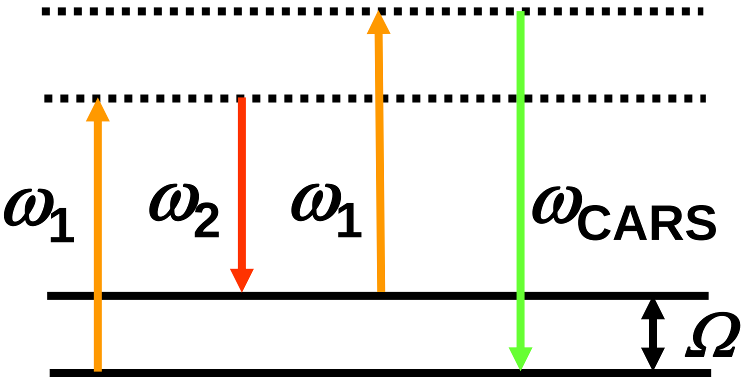
Sym. species	No.	Approximate type of mode	Selected value of frequency	Infrared	Raman	Comments
				cm^{-1} (Gas)	cm^{-1} (Liquid)	
a_g	ν_1	CH_2 s-stretch	2957 D	ia	2957 (10) p	
	ν_2	CH_2 scis	1445 C	ia	1445 (4b) dp	
	ν_3	CH_2 wag	1304 C	ia	1304 (6) p	
	ν_4	CC stretch	1052 C	ia	1052 (4) p	
	ν_5	CCl stretch	754 C	ia	754 (10b) p	
	ν_6	CCCl deform	300 C	ia	300 (8) p	
a_u	ν_7	CH_2 a-stretch	3005 D	3005 W (liquid)	ia	SF (gauche ν_1, gauche ν_{11}).
	ν_8	CH_2 twist	1123 B	1122.5 W	ia	
	ν_9	CH_2 rock	773 B	772.5 M	ia	
	ν_{10}	Torsion	123 C	123 M	ia	
b_g	ν_{11}	CH_2 a-stretch	3005 D	ia	3005 (8b) dp	
	ν_{12}	CH_2 twist	1264 C	ia	1264 (3) dp	
	ν_{13}	CH_2 rock	989 C	ia	989 (2) p	
b_u	ν_{14}	CH_2 s-stretch	2983 C	2983.3 M	ia	
	ν_{15}	CH_2 scis	1461 A	1460.6 S	ia	
	ν_{16}	CH_2 wag	1232 B	1232.3 S	ia	
	ν_{17}	CCl stretch	728 C	728.3 VS	ia	
	ν_{18}	CCCl deform	222 C	222.3 W	ia	

T. Shimanouchi, Tables of Molecular Vibrational Frequencies, NSRDS-NBS 39, p. 97.

Polarization-resolved CARS Spectroscopy

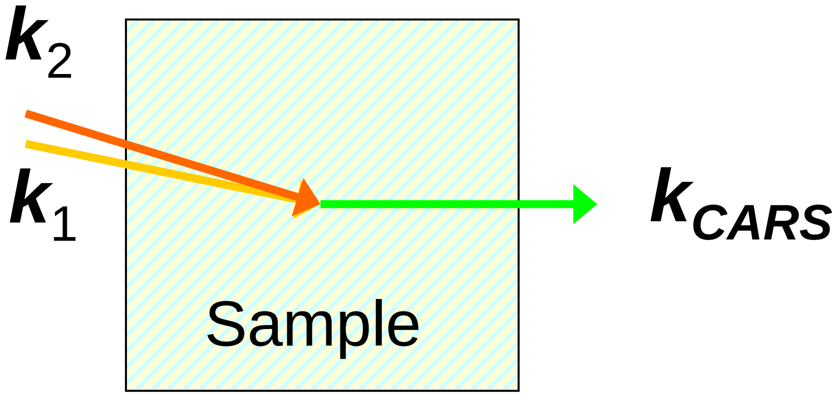
Energy conservation:

$\omega_{\mathrm{CARS}} = 2\omega_1 - \omega_2 = \omega_1 + \Omega$

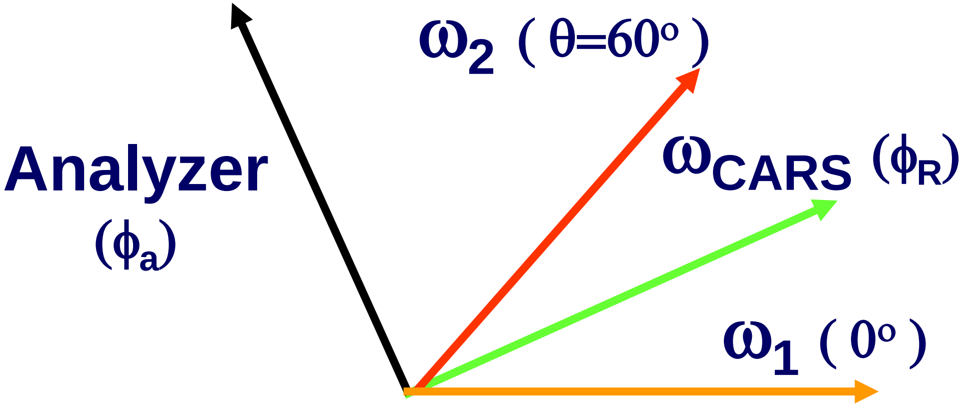


Momentum conservation:

$\boldsymbol{k} = 2\boldsymbol{k}_1 - \boldsymbol{k}_2$



Polarization rule: $\tan\phi_R = \rho \tan\theta$ (ρ; Raman depolarization ratio)



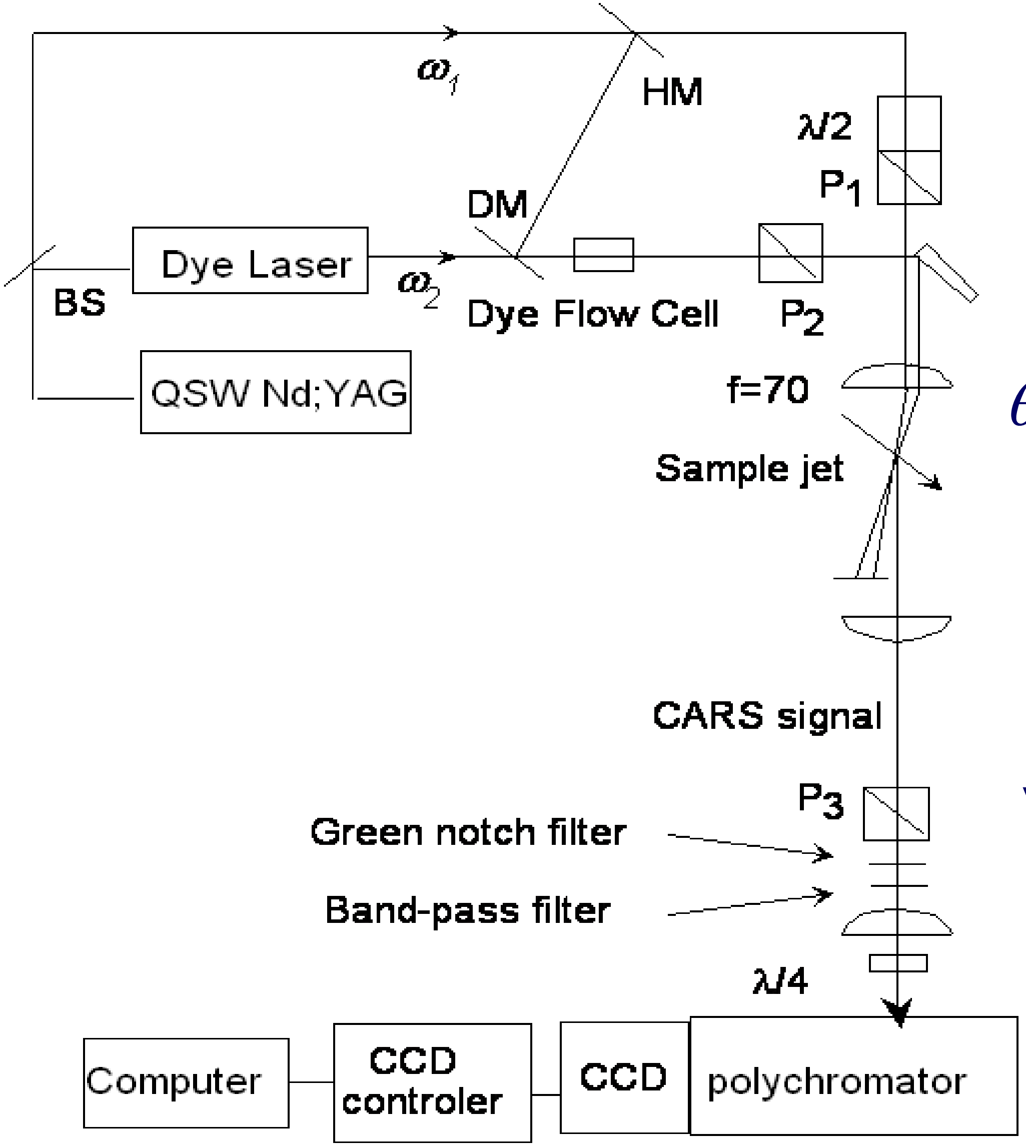
p: $\rho<0.75$

Totally symmetric mode

dp: $\rho=0.75$

Non-totally symmetric mode

Polarization-resolved CARS Spectroscopy



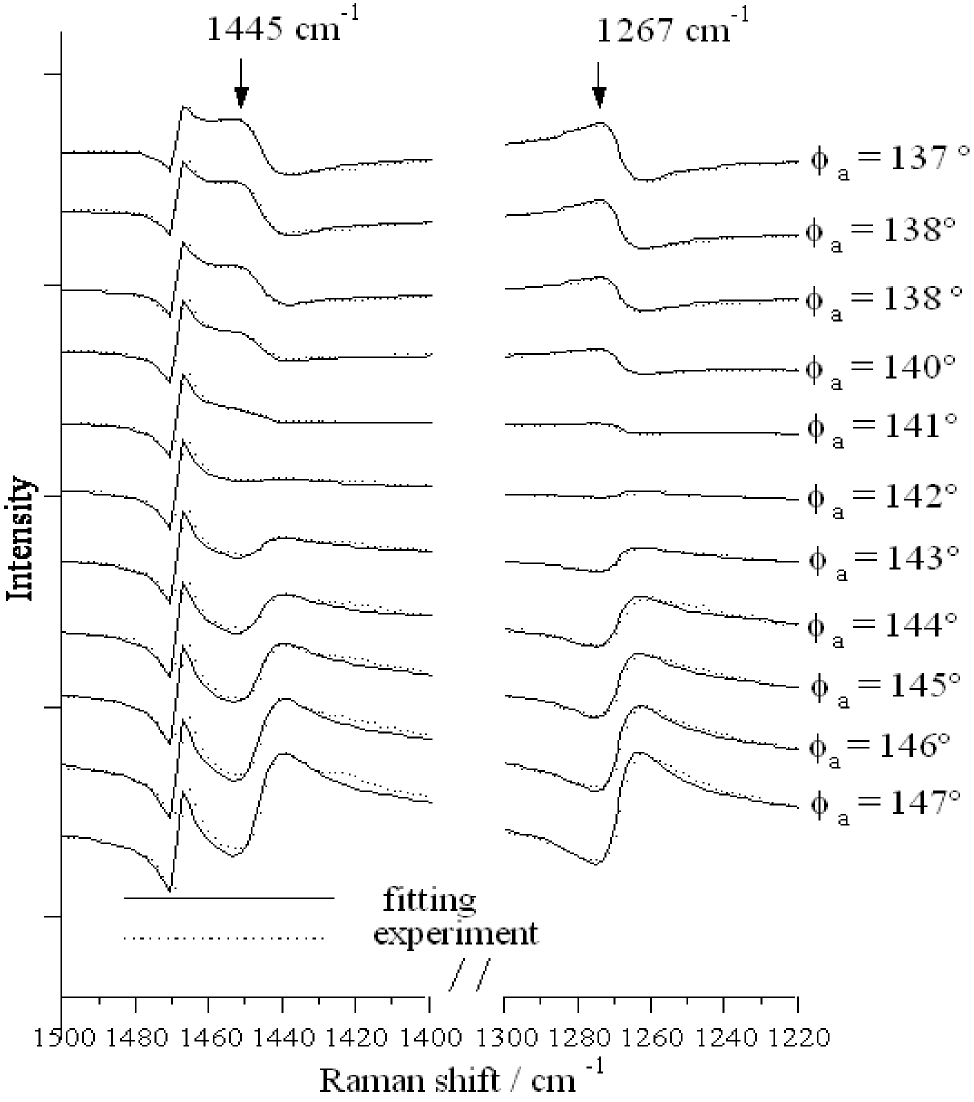
$\theta = 60^o$

Variable ϕ_a

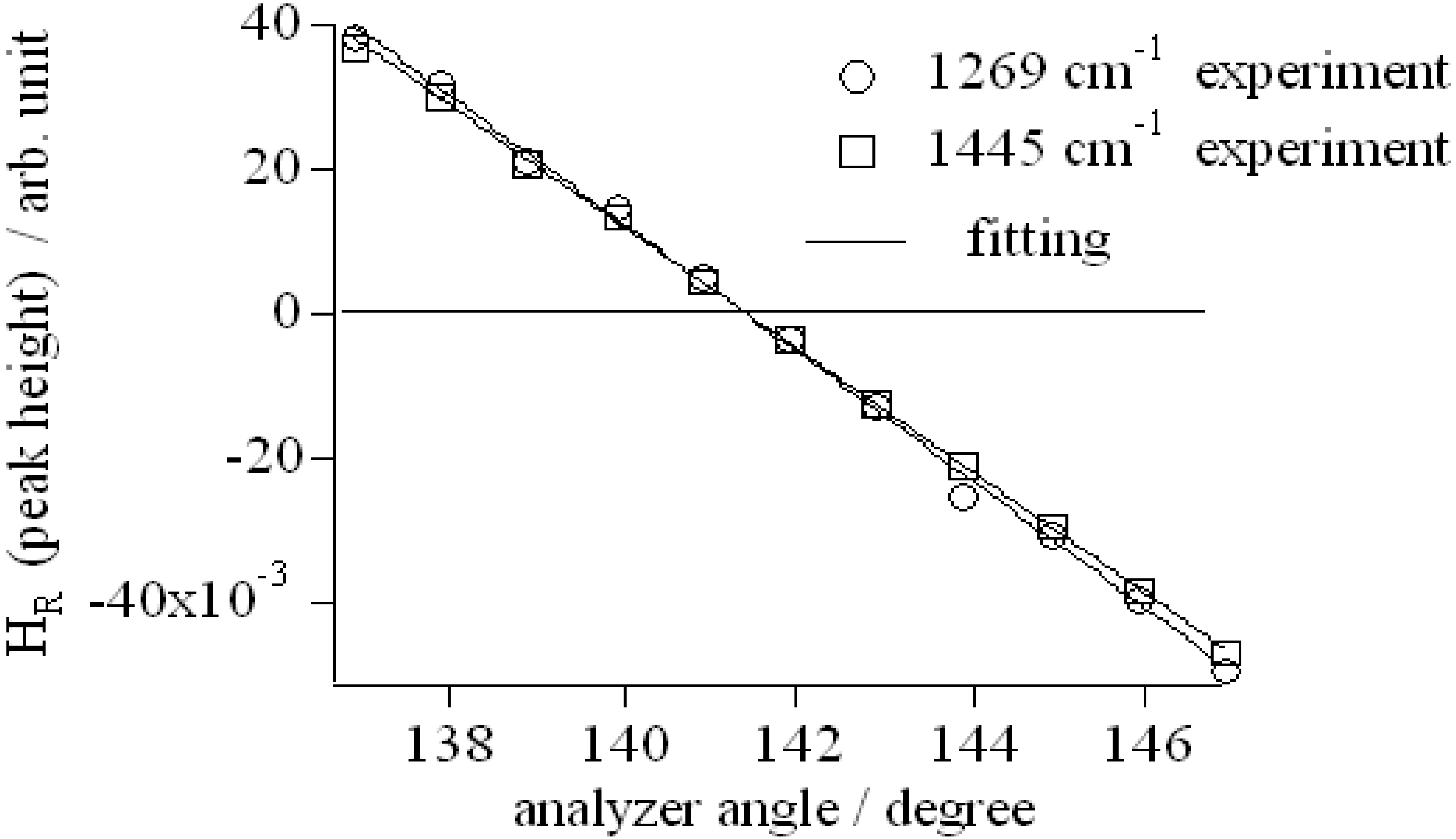
Y. Saito, T. Ishibashi, H. Hamaguchi, *J. Raman Spectrosc.*, **31**, 725-730 (2000).

Polarization-resolved CARS Spectra of Liquid Cyclohexane

$$\mathbf{I}_{\mathbf{CARS}} = \left(A_{NR} + \sum_{R} \frac{H_R \mathrm{G}_\mathrm{R}}{\mathrm{w}_R - (\mathrm{w}_1 - \mathrm{w}_2) - i\mathrm{G}_\mathrm{R}} \right)^2$$

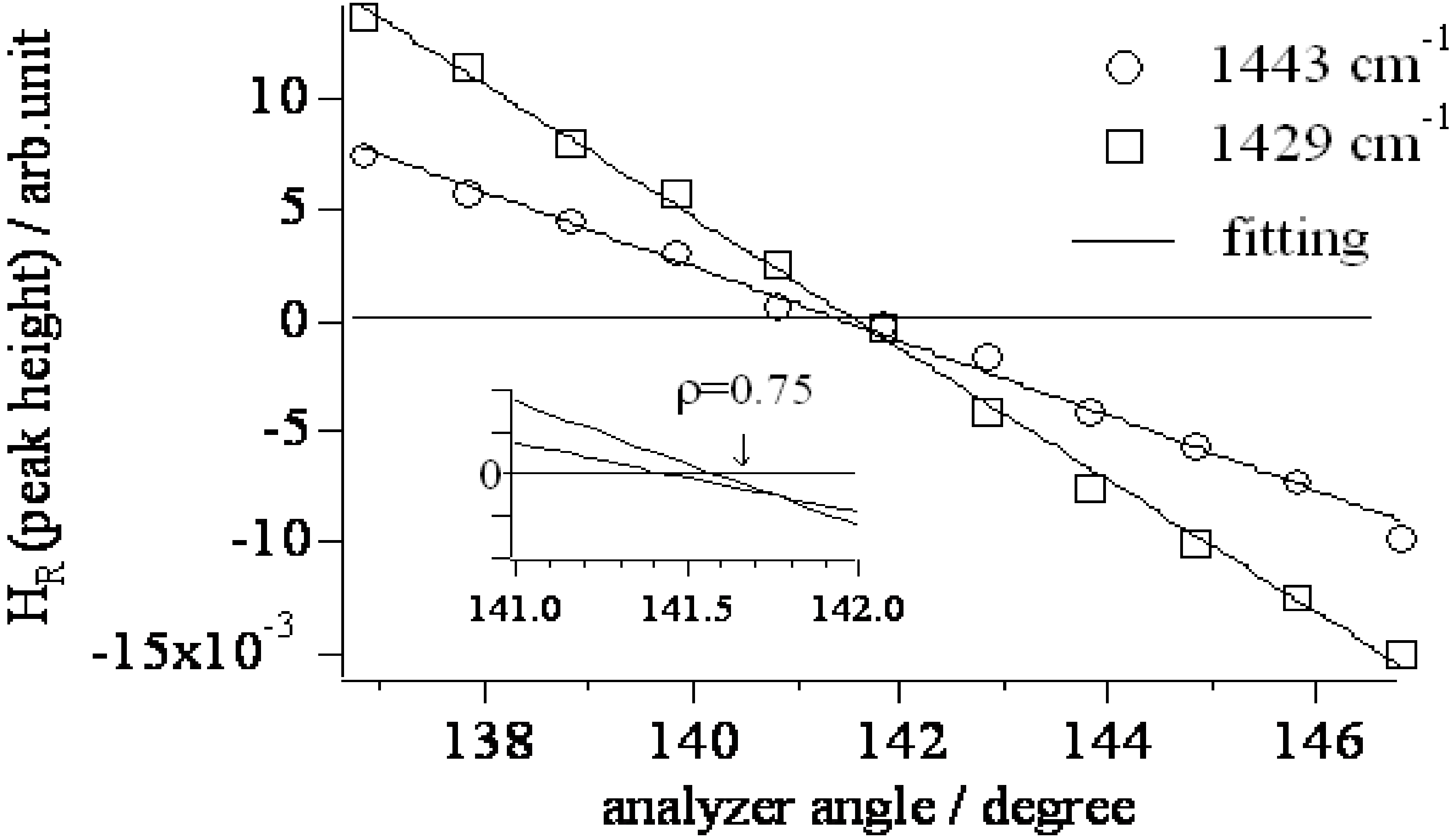


Depolarization Ratios of Two e_g Bands of Cyclohexane



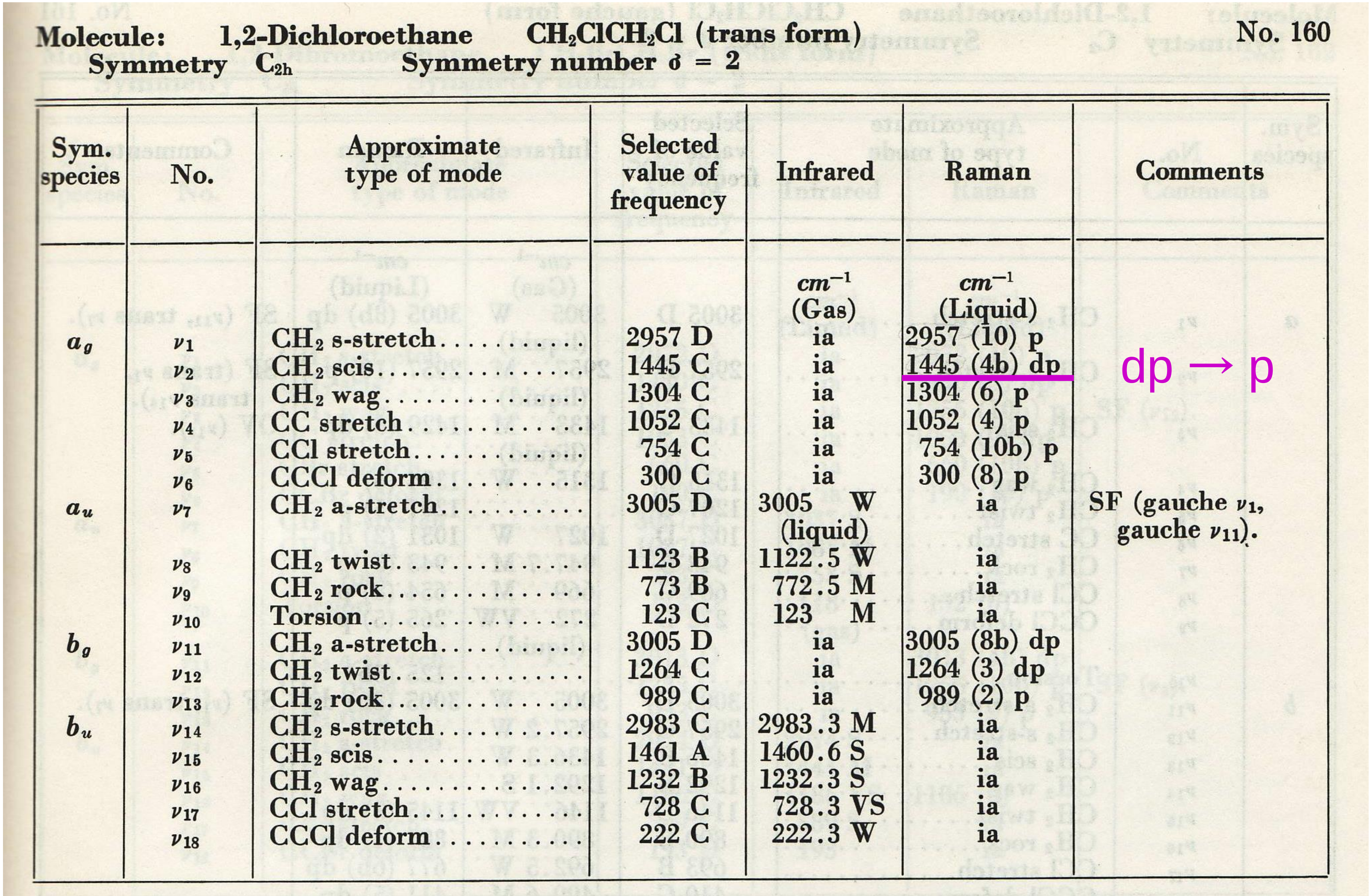
CH_2 twist	(e_g)	1267 cm^{-1}	0.749±0.002
CH_2 scissors	(e_g)	1445 cm^{-1}	0.750±0.002

Depolarization Ratios of Two Totally-symmetric Bands of 1,2-dichloroethane



gauche CH_2 scissors (a)	1429 cm $^{-1}$	0.746±0.003
trans CH_2 scissors (a_g)	1443 cm $^{-1}$	0.742±0.003

Depolarized Totally-symmetric Raman Band?



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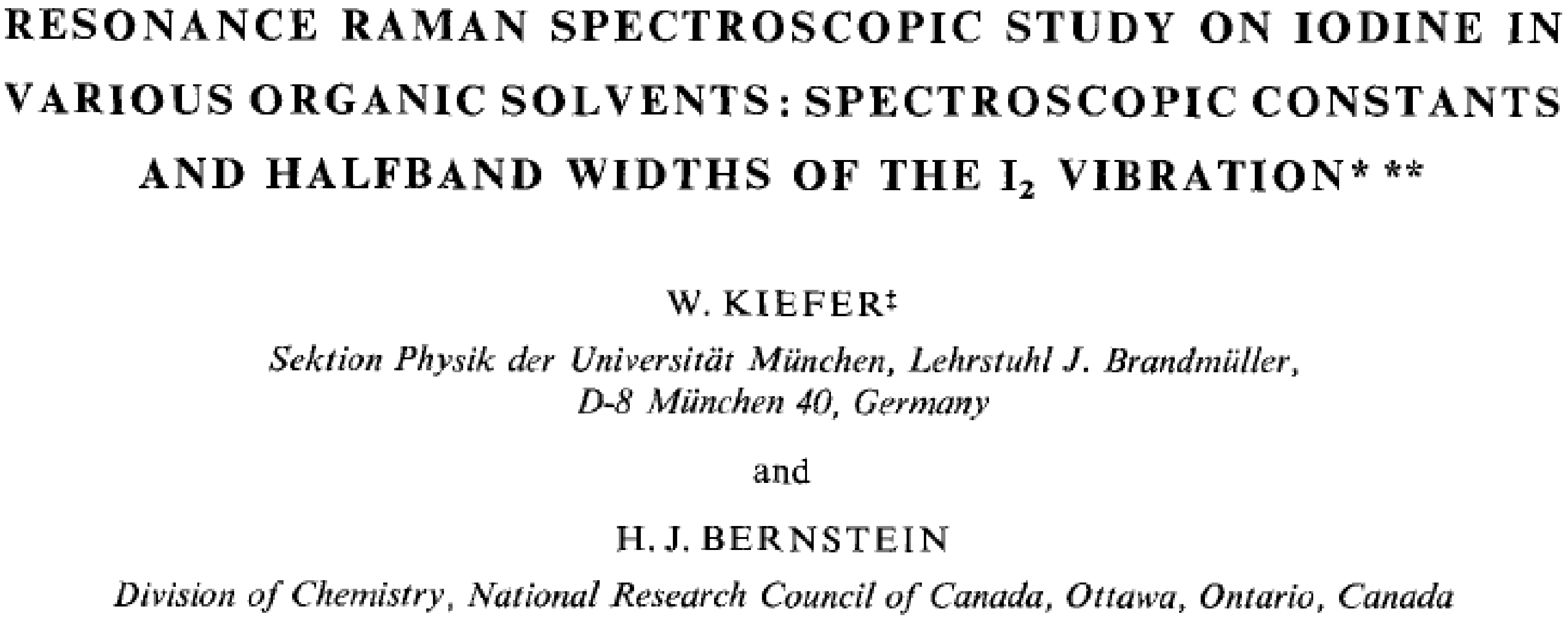
dp → p

T. Shimanouchi, Tables of Molecular Vibrational Frequencies, NSRDS-NBS 39, p. 97.

Resonance Raman scattering



RESONANCE RAMAN SPECTROSCOPIC STUDY ON IODINE IN VARIOUS ORGANIC SOLVENTS: SPECTROSCOPIC CONSTANTS AND HALFBAND WIDTHS OF THE I_2 VIBRATION* **

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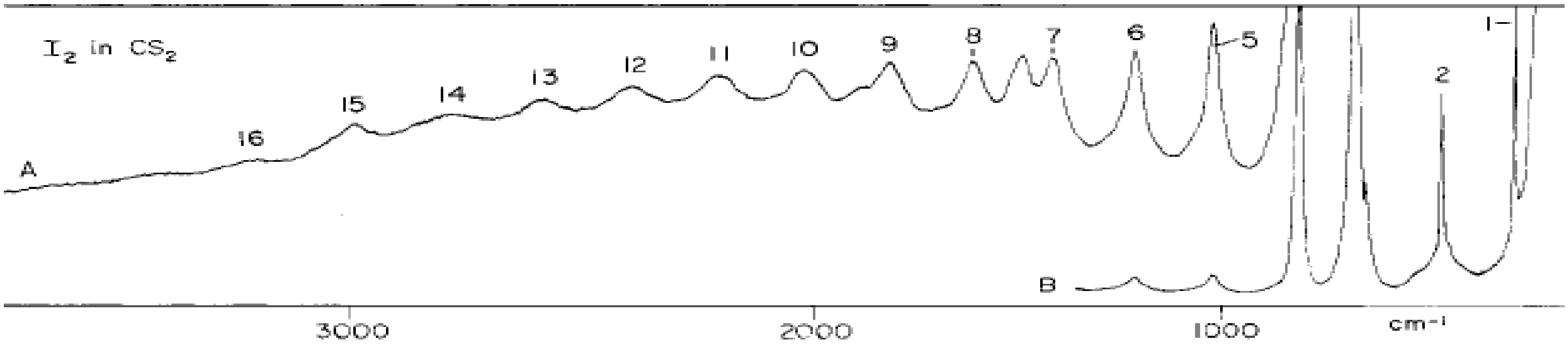
and

H. J. BERNSTEIN
Division of Chemistry, National Research Council of Canada, Ottawa, Ontario, Canada



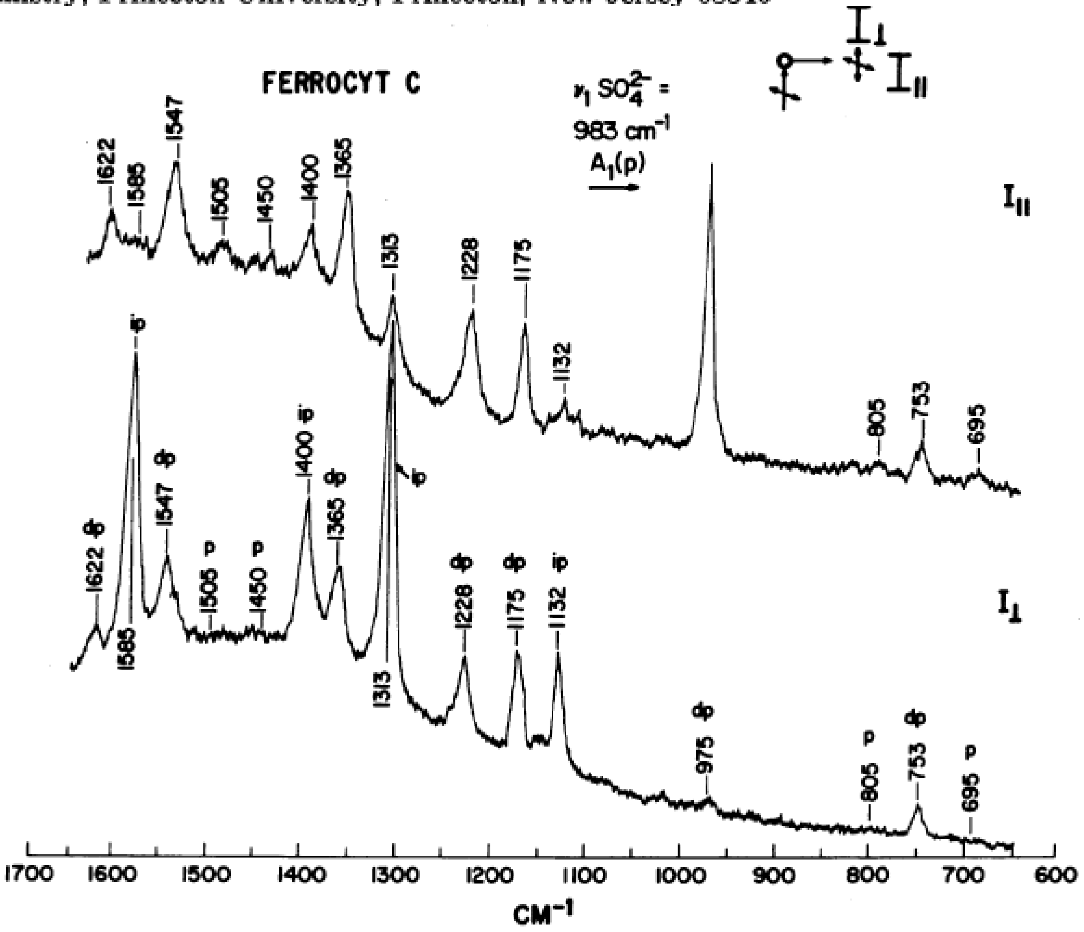
Proc. Nat. Acad. Sci. USA
Vol. 69, No. 9, pp. 2622–2626, September 1972

Resonance Raman Spectra of Hemoglobin and Cytochrome *c*: Inverse Polarization and Vibronic Scattering

(scattering tensor/porphyrin fluorescence)

THOMAS G. SPIRO AND THOMAS C. STREKAS

Department of Chemistry, Princeton University, Princeton, New Jersey 08540



Theoretical Framework of Raman Spectroscopy

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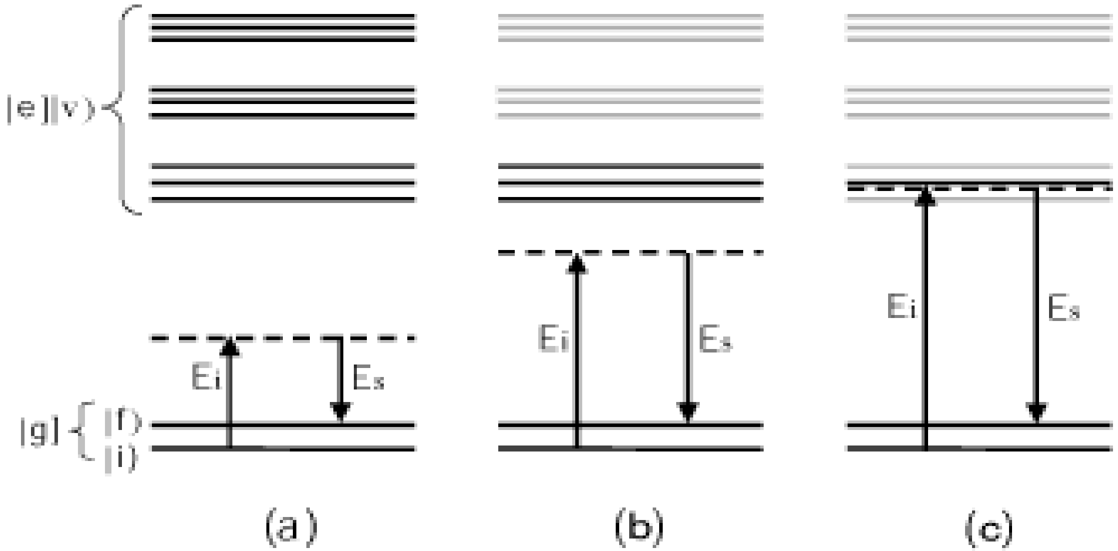
Vibrational Raman scattering and electronic resonance

In vibrational Raman scattering in the ground electronic state, the initial, final and intermediate states are expressed as the products of the electronic and vibrational parts as,

|m >=|g]|i)
|n>=|g]|f)
|e>=|e]|v)



where |] stands for electronic state and |) for vibrational state; |g] is the ground electronic state and |e] the excited electronic state(s), and |i), |f),|v) are the initial, final and intermediate vibrational states, respectively.

In off-resonance Raman scattering (a), E_{ev}-E_{gi}» Ei and therefore many excited electronic states Σ |e])|v) contribute (virtual intermediate states). In pre-resonance Raman scattering, Ei become close to E_{ev}-E_{gi} and the vibrational states of the lowest excited electronic state |e]Σ|v) play the role of interemediate states. In rigorous resonance Raman scattering, E_{ev}-E_{gi}~ Ei and one particular vibronic state |e]|v) dominates the scattering process.

For resonance Raman scattering, we need to take explicitly the vibronic nature of the intermediate states. We write the molecular Hamiltonian with three terms, electronoc, vibrational and vibronic terms..

$$H_{mol}=H_{mol}^{e}+ \mathrm{H}_{mol}^{v} +\mathrm{H}_{mol}^{ev} \tag{41}$$

We treat H_{mol}^{ev} as a perturbation to H_{mol}^{e} and H_{mol}^{v}. We consider three zero-order electronic states, ground electronic state $|g_0]$, the resonant excited electronic state $|e_0]$ and another excited electronic state $|s_0]$. They satisfy the following equations.

$$H_{mol}^{e}|g_0]= E_g^0|g_0] \tag{42}$$

$$H_{mol}^{e}|e_0]= E_e^0|e_0] \tag{43}$$

$$H_{mol}^{e}|s_0]= E_s^0|s_0] \tag{44}$$

Three vibrational states are the initial vibrational state |i), intermediate vibrational state |v) and the final vibrational state |f).

$$\mathrm{H}_{mol}^{v}|i)= E_i^0|i) \tag{45}$$

$$\mathrm{H}_{mol}^{v}|v)= E_v^0|v) \tag{46}$$

$$\mathrm{H}_{mol}^{v}|f)= E_f^0|f) \tag{47}$$

With the first-order perturbation theory, we obtain an expression for the first-order intermediate electronic state (Herzberg-Teller expansion)

$$|e] = \sum_{s \neq e,g} [s_0 | \left(\frac{\partial H_{mol}^{ev}}{\partial Q_k} \right)_0 Q_k | e_0] | s_0]$$

Introducing all into the KHD formula, we obtain an expression for a Raman scattering tensor under a resonance condition (Albrecht 1961).

$$\alpha_{\rho\sigma} = \mathrm{A} + \mathrm{B}$$

$$\mathrm{A} = \sum_{e \neq g} \sum_{v} \frac{[g_0 | D_\sigma | e_0] | [e_0 | D_\rho | g_0]}{E_e^0 + E_v^0 - E_g^0 - E_i^0 - E_i - i\Gamma_e} (i | v)(v | f)$$

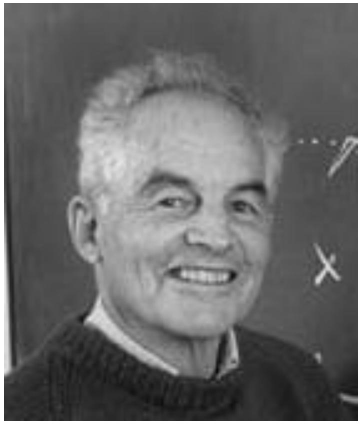
$$\mathrm{B} = \sum_{s \neq g} \sum_{v} \sum_{l} \frac{[g_0 | D_\sigma | s_0][s_0 | (\partial H_{ev} / \partial Q_l)_0 | e_0][e_0 | D_\rho | g_0]}{E_s^0 + E_v^0 - E_g^0 - E_i^0 - E_i - i\Gamma_s} (i | Q_l | v)(v | f)$$

$$+ \sum_{s \neq g} \sum_{v} \sum_{l} \frac{[g_0 | D_\sigma | e_0][e_0 | (\partial H_{ev} / \partial Q_l)_0 | s_0][s_0 | D_\rho | g_0]}{E_s^0 + E_v^0 - E_g^0 - E_i^0 - E_i - i\Gamma_s} (i | v)(v | Q_l | f)$$

Theory of Raman Scattering (3)

Albrecht's vibronic theory of resonance Raman Scattering

A. C. Albrecht, J. Chem. Phys. 34, 1476 (1961).



A. C. Albrecht
(1927-2002)

$$a_{\rho\sigma} \sim A + B$$

A term: Franck-Condon term

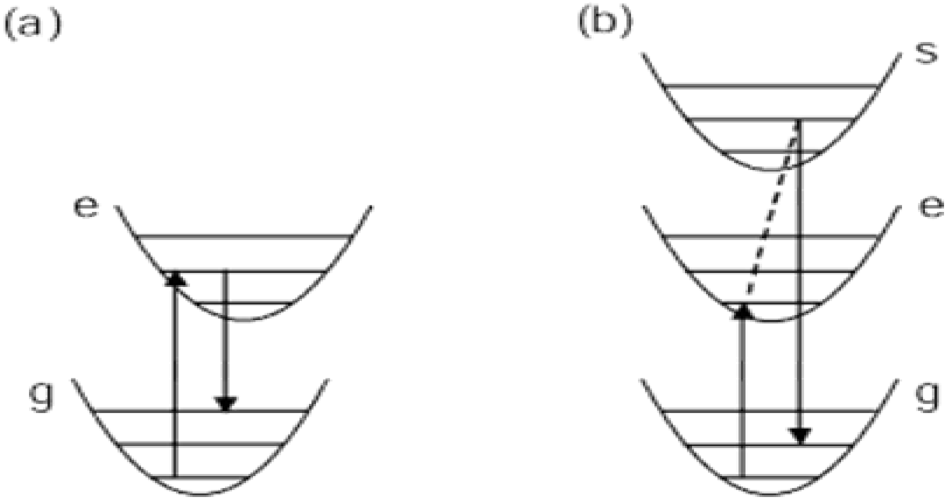
Totally symmetric modes

high overtones

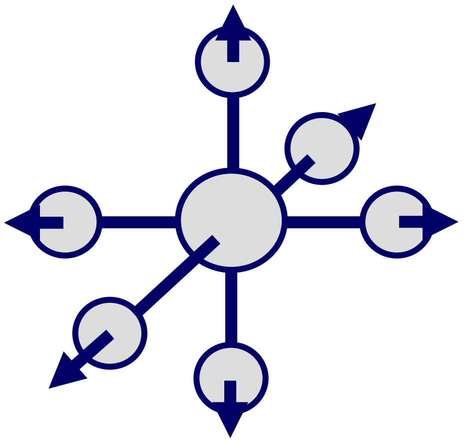
B term: Vibronic coupling

Non-totally symmetric modes

overtones?



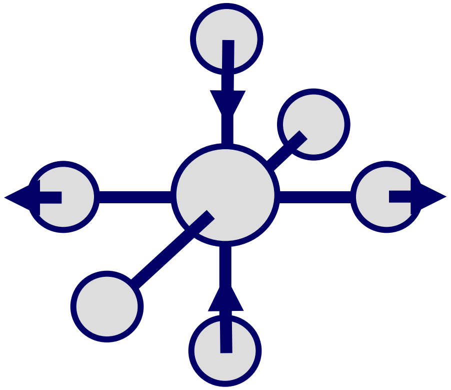
Raman Active Vibrations of MX_6 Octahedral Complexes



ν_1
(a_{1g})

$$a_{1g} \sim \begin{pmatrix} 1 & 0 & 0 \\ 0 & 1 & 0 \\ 0 & 0 & 1 \end{pmatrix}$$

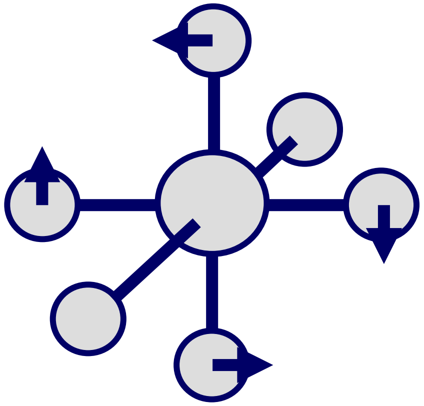
$\rho=0$



ν_2
(e_g)

$$e_g \sim \begin{pmatrix} 1 & 0 & 0 \\ 0 & -1 & 0 \\ 0 & 0 & 0 \end{pmatrix}$$

$\rho=0.75$

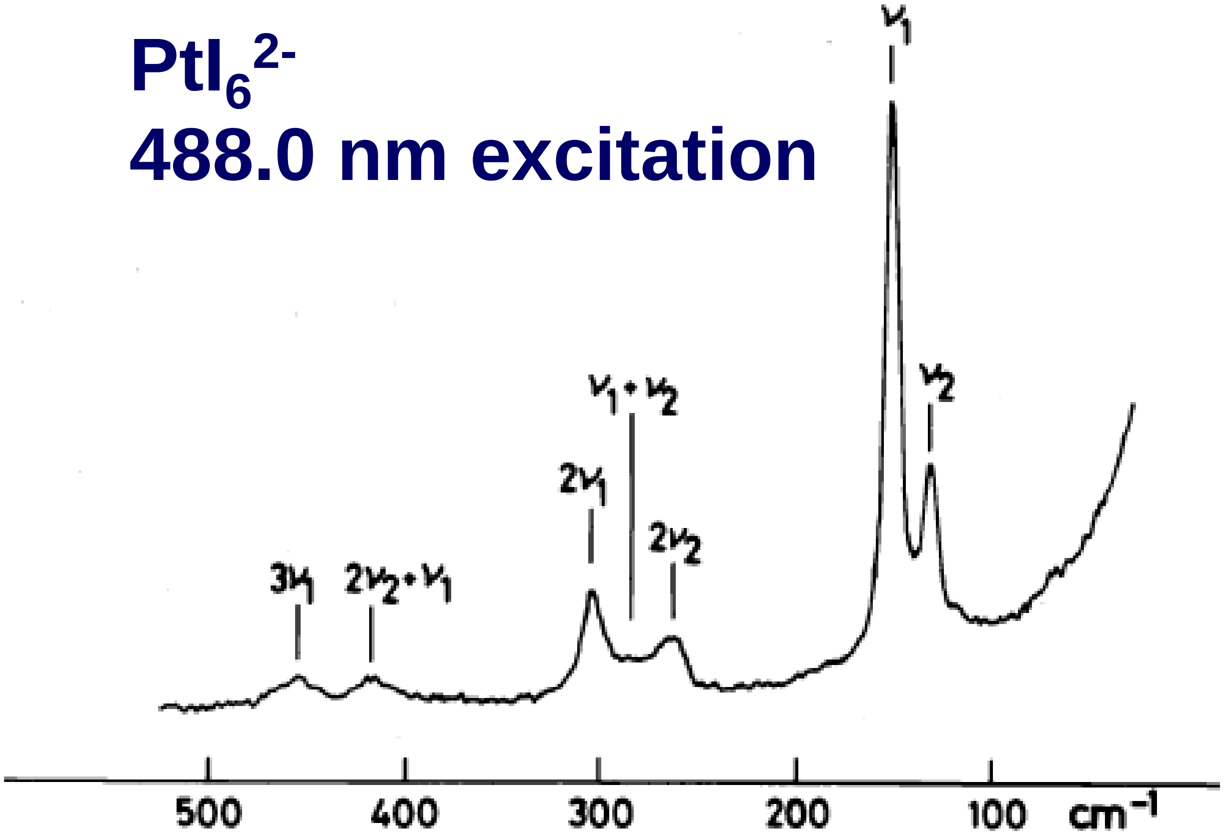


ν_5
(t_{2g})

$$t_{2g} \sim \begin{pmatrix} 0 & 1 & 0 \\ 1 & 0 & 0 \\ 0 & 0 & 0 \end{pmatrix}$$

$\rho=0.75$

Resonance Raman spectrum of PtI_6^{2-}



PtI_6^{2-}

488.0 nm excitation

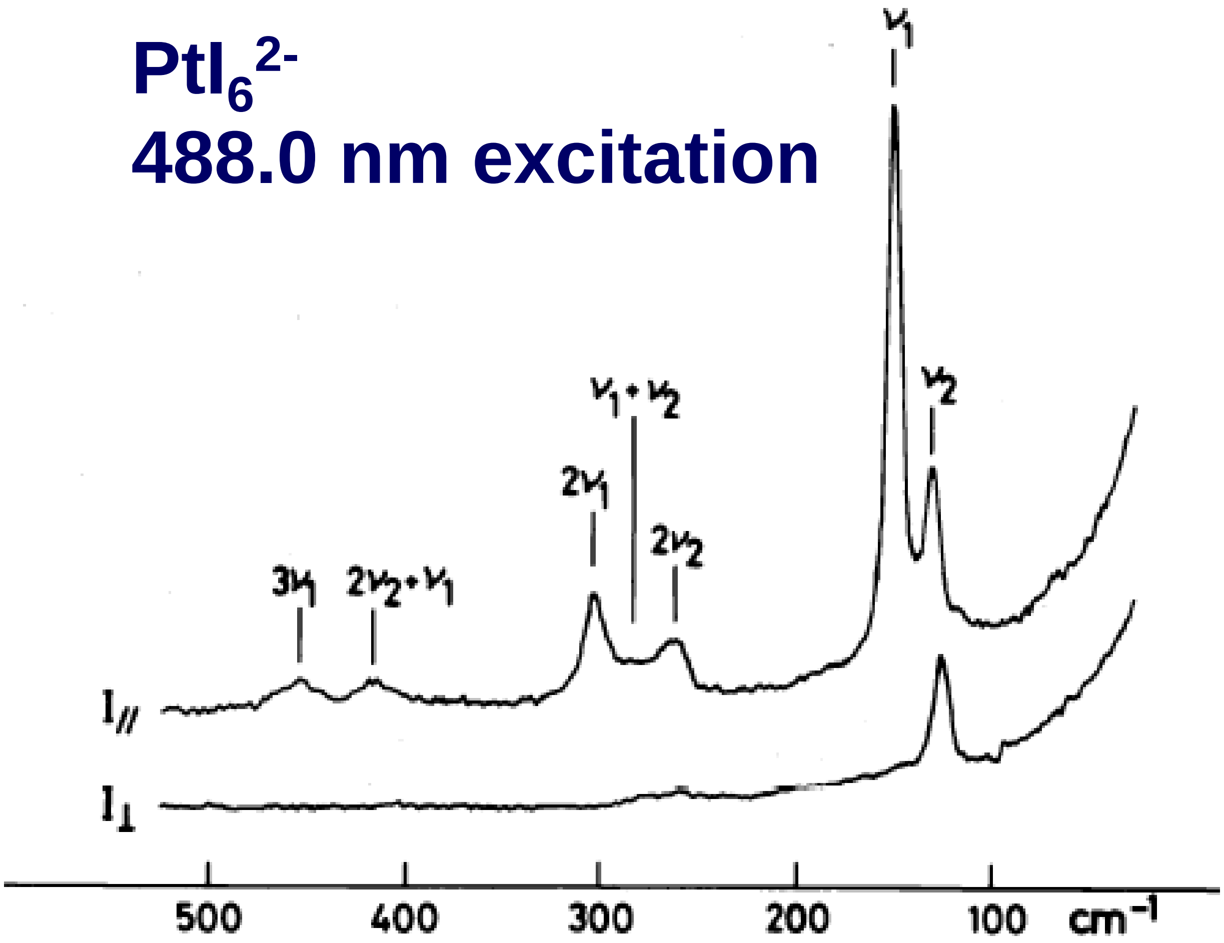
Totally symmetric mode （ν_1, $2\nu_1$, $3\nu_1$） → **A term**

Non-totally symmetric mode （ν_2, $2\nu_2$, $\nu_1+\nu_2$, $2\nu_1+\nu_2$） → **B term**

H. Hamaguchi, I. Harada, T. Shimanouch, *J. Raman Spectrosc.*, **2**, 517-528 (1974).

Polarized Resonance Raman Spectra of PtI_6^{2-}

PtI_6^{2-}
488.0 nm excitation

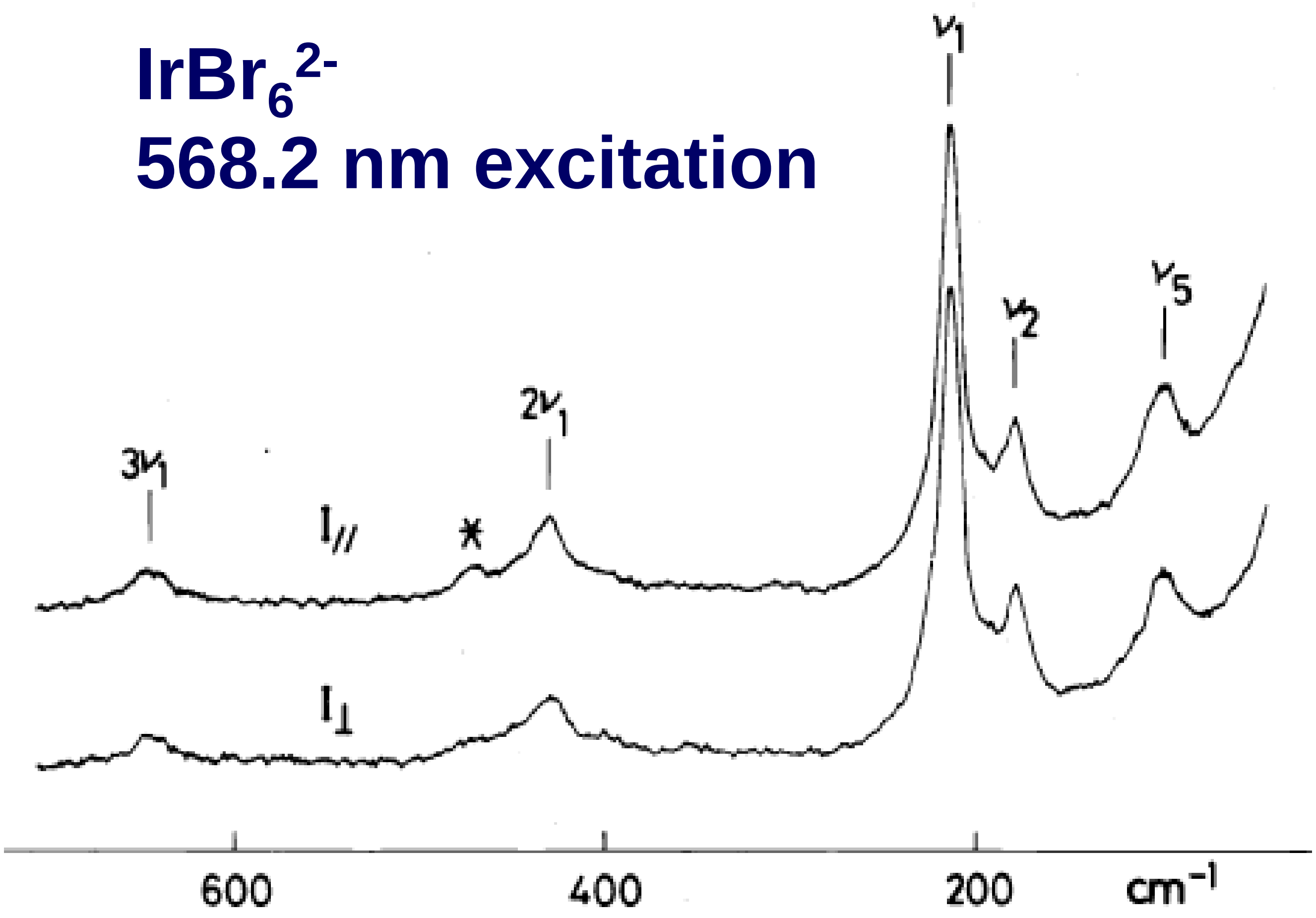


ν_1, $2\nu_1$, $3\nu_1$ bands $\rho=0$; ν_2 band $\rho=0.75$

H. Hamaguchi, *J. Chem. Phys.*, **69**, 569-578 (1978).

Polarized Resonance Raman Spectra of $IrBr_6^{2-}$

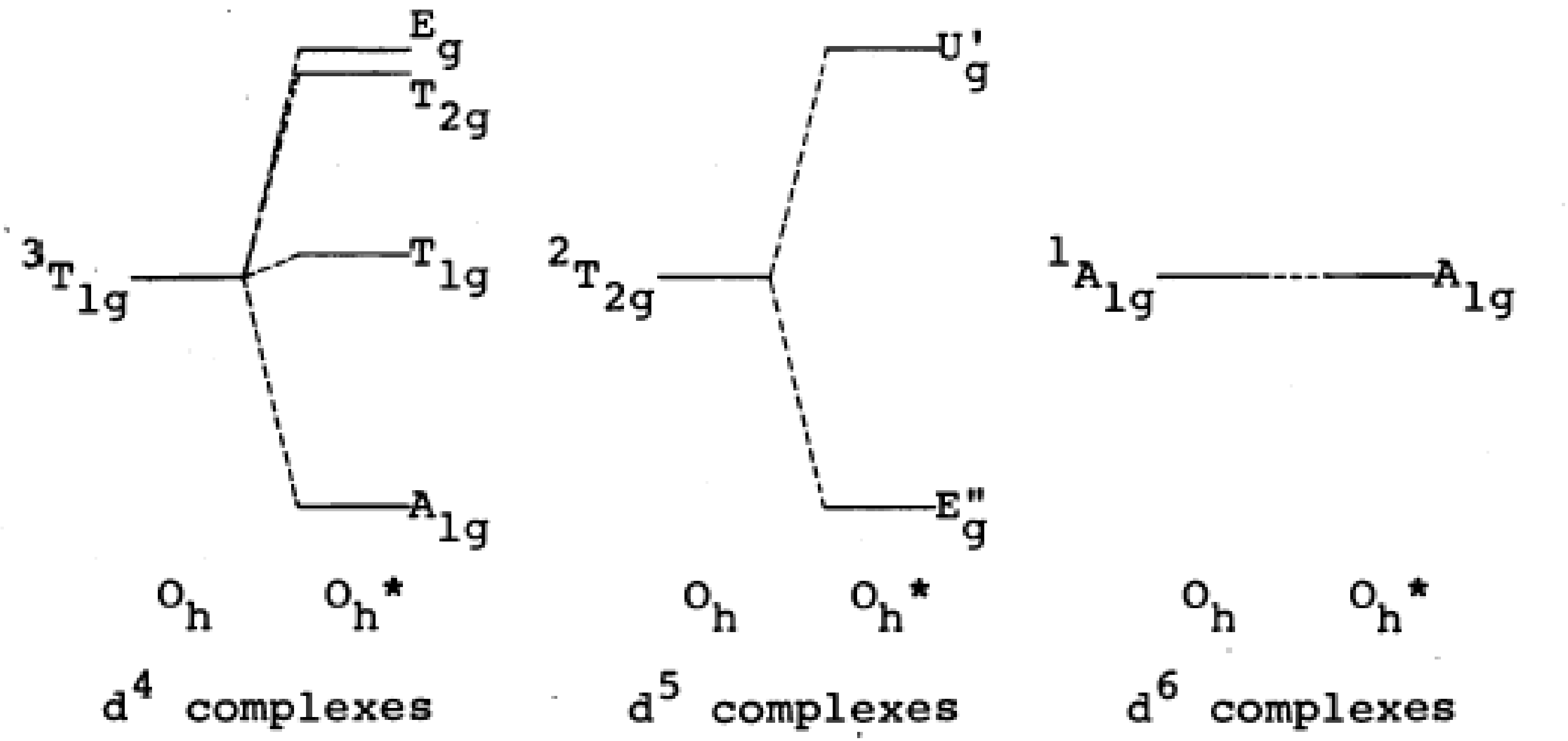
$IrBr_6^{2-}$
568.2 nm excitation



ρ=1 for all bands; forgot to rotate the analyzer?

H. Hamaguchi, *J. Chem. Phys.*, **66**, 5757-5768 (1977)； **69**, 569-578 (1978).

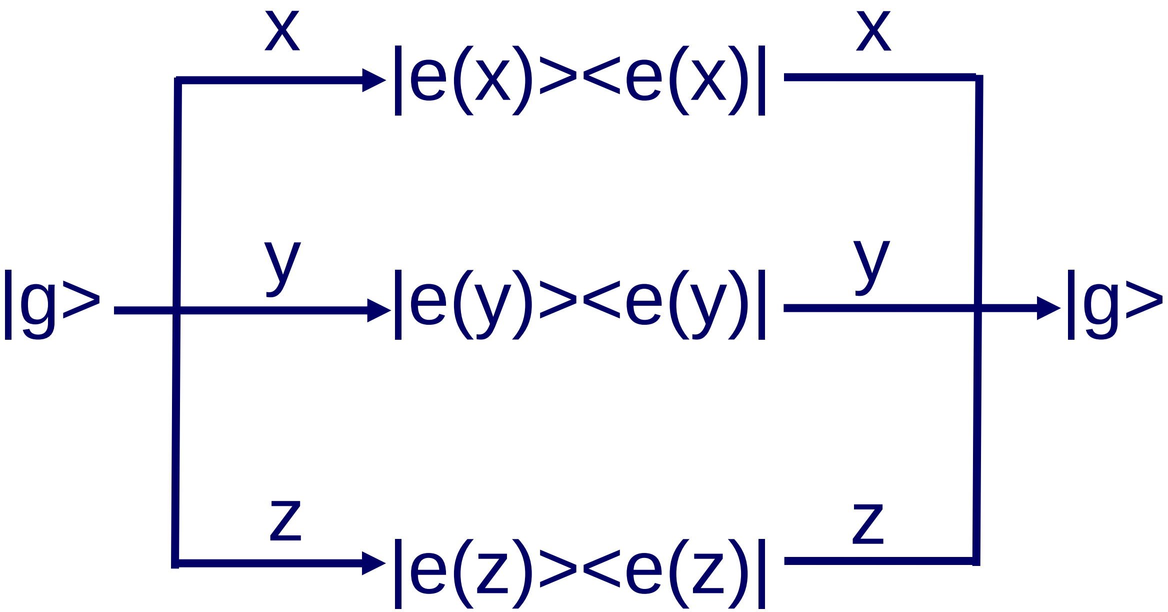
Ground-state Electron Configuration and Electronic States of Octahedral MX_6 Complexes



$OsCl_6^{2-}$ **$IrBr_6^{2-}$** **PtI_6^{2-}**

Raman Scattering Tensor and Depolarization Ratio of the Totally Symmetric Mode of Non-degenerate PtI_6^{2-} Ion

$\Gamma_g = a_{1g}$, $\Gamma_e = t_{1u}$



x x

$|e(x)\rangle\langle e(x)|$

$|g\rangle$ y y $|g\rangle$

$|e(y)\rangle\langle e(y)|$

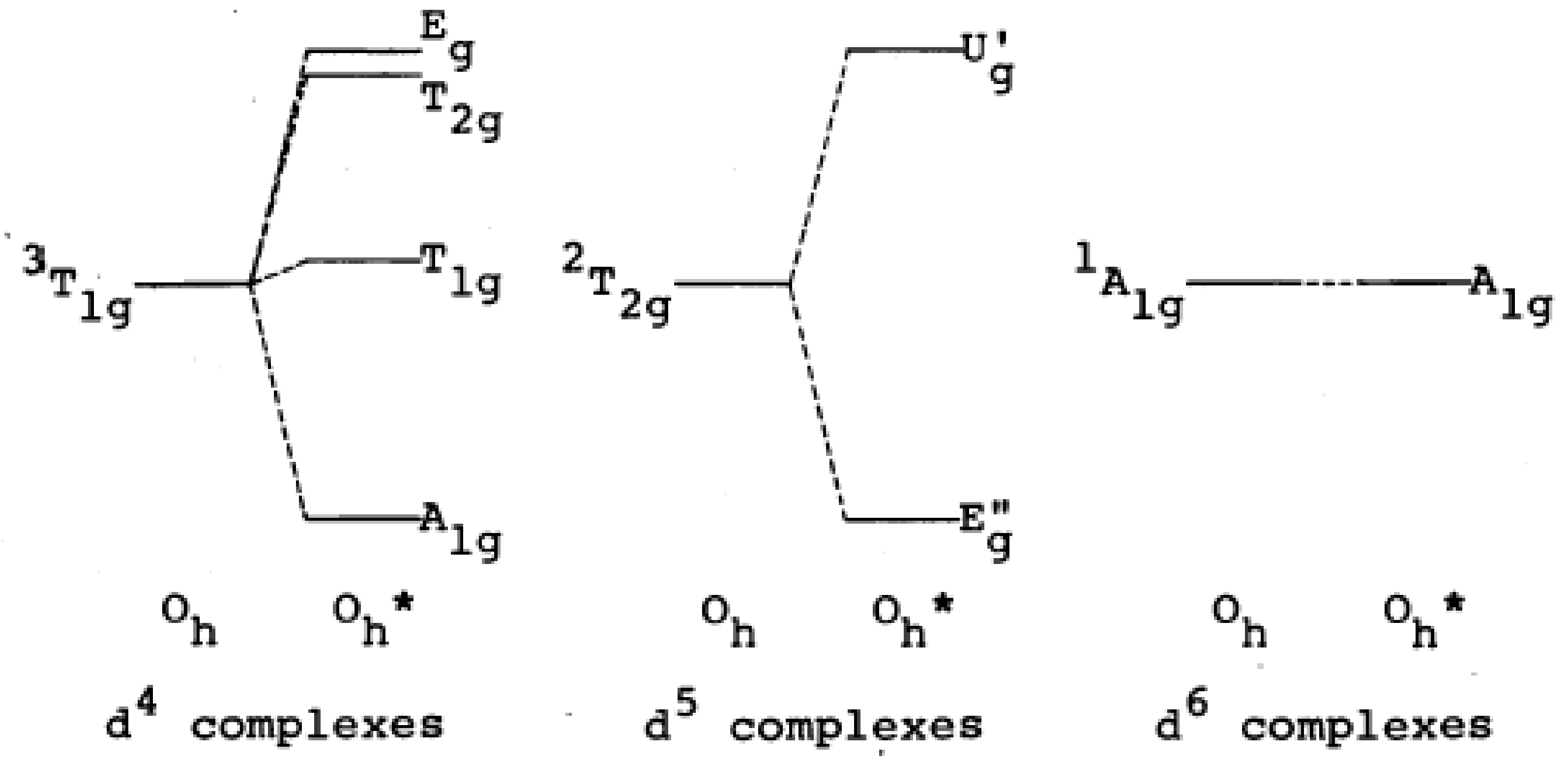
z z

$|e(z)\rangle\langle e(z)|$

$$\begin{pmatrix}1 & 0 & 0\\ 0 & 1 & 0\\ 0 & 0 & 1\end{pmatrix}$$

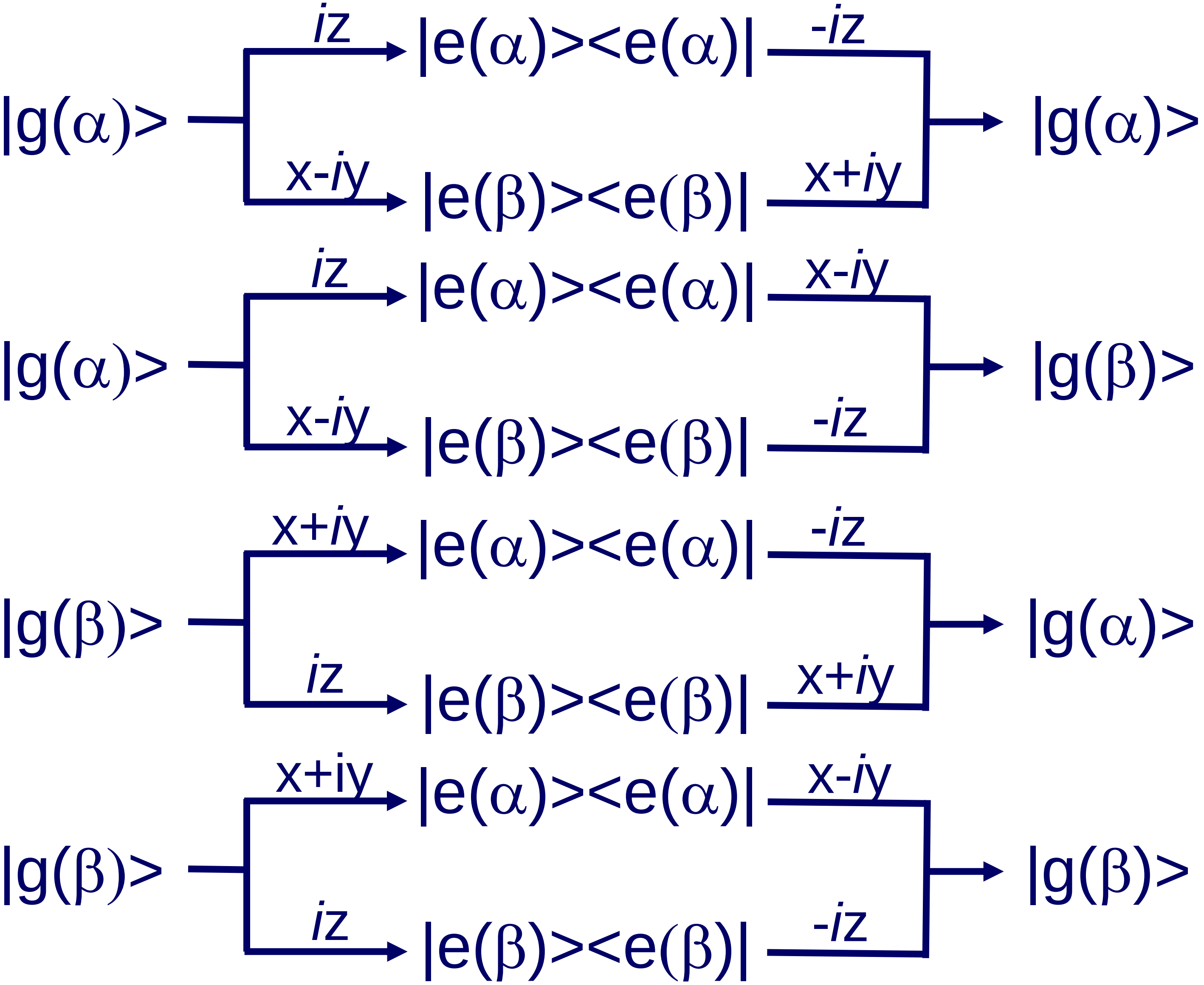
$G_0=3$, $G_a=0$, $G_s=0$ $\rho=3G_s/(10G_0+4G_s)=0$

Ground-state Electron Configuration and Electronic States of Octahedral MX_6 Complexes



$OsCl_6^{2-}$ **$IrBr_6^{2-}$** **PtI_6^{2-}**

Raman Tensors for the a_{1g} Vibrational Transitions of MX_6 in an E_g” Ground Electronic State



|g(α)> *iz* |e(α)><e(α)| -*iz* x-*i*y |e(β)><e(β)| x+*i*y |g(α)>

$$\begin{bmatrix} 1 & i & 0 \\ -i & 1 & 0 \\ 0 & 0 & 1 \end{bmatrix}$$

|g(α)> *iz* |e(α)><e(α)| x-*i*y x-*i*y |e(β)><e(β)| -*iz* |g(β)>

$$\begin{bmatrix} 0 & 0 & -i \\ 0 & 0 & -1 \\ i & 1 & 0 \end{bmatrix}$$

|g(β)> x+*i*y |e(α)><e(α)| -*iz* *iz* |e(β)><e(β)| x+*i*y |g(α)>

$$\begin{bmatrix} 0 & 0 & -i \\ 0 & 0 & 1 \\ i & -1 & 0 \end{bmatrix}$$

|g(β)> x+iy |e(α)><e(α)| x-*i*y *iz* |e(β)><e(β)| -*iz* |g(β)>

$$\begin{bmatrix} 1 & -i & 0 \\ i & 1 & 0 \\ 0 & 0 & 1 \end{bmatrix}$$

Raman Tensors for the a_{1g} Vibrational Transitions of MX_6 in an E_g” Ground Electronic State and Depolarization Ratio

$$|g(\alpha)> \longrightarrow |g(\alpha)> \qquad \begin{pmatrix} 1 & i & 0 \\ -i & 1 & 0 \\ 0 & 0 & 1 \end{pmatrix} \qquad G_0=3,\ G_a=2,\ G_s=0$$

$$|g(\alpha)> \longrightarrow |g(\beta)> \qquad \begin{pmatrix} 0 & 0 & -i \\ 0 & 0 & -1 \\ i & 1 & 0 \end{pmatrix} \qquad G_0=0,\ G_a=4,\ G_s=0$$

$$|g(\beta)> \longrightarrow |g(\alpha)> \qquad \begin{pmatrix} 0 & 0 & -i \\ 0 & 0 & 1 \\ i & -1 & 0 \end{pmatrix} \qquad G_0=0,\ G_a=4,\ G_s=0$$

$$|g(\beta)> \longrightarrow |g(\beta)> \qquad \begin{pmatrix} 1 & -i & 0 \\ i & 1 & 0 \\ 0 & 0 & 1 \end{pmatrix} \qquad G_0=3,\ G_a=2,\ G_s=0$$

$$G_0=6,\ G_a=12,\ G_s=0 \qquad \rho=(3G_s+5G_a)/(10G_0+4G_s)=1$$

Symmetry of Raman Scattering Tensor

Irreducible representations:

Γ_i: the initial states, Γ_f: the final state, Γ_R: Raman tensor

$$\Gamma_R = \Gamma_i \times \Gamma_f$$

Vibrational Raman Scattering:

$\Gamma_i = \gamma_g \times \gamma_1$, $\Gamma_f = \gamma_g \times \gamma_v$ and therefore $\Gamma_R = \gamma_g \times \gamma_g \times \gamma_v$

where γ_g: the ground electronic state and γ_v: vibrational state

A_{1g} Vibrational Raman Scattering:

d_6 PtI_6^{2-} $\gamma_g = a_{1g}$, $\gamma_v = a_{1g}$ and therefore $\Gamma_R = a_{1g}$ $\rho = 0$

d_5 $IrBr_6^{2-}$ $\gamma_g = e_g$" $\gamma_v = a_{1g}$ and therefore $\gamma_R = a_{1g} + t_{1g}$ $0 < r < \infty$

Depolarization Ratio in Vibrational Raman Scattering

Polarizability theory (Placzek, 1934)

1) Non-resonant condition, 2) Non-degenerate condition

Totally symmetric modes: $G_0 \neq 0$, $G_a = 0$, $G_s \neq 0 \Longrightarrow$ $0 \leq \rho < 0.75$

Non-totally symmetric modes: $G_0 = 0$, $G_a = 0$, $G_s \neq 0 \Longrightarrow$ $\rho = 0.75$

Breakdown of the Placzek polarizability theory

1) Non-resonant condition (Spiro, 1972)

Non-totally symmetric modes : $G0 = 0$, $G_a \neq 0$, $Gs \neq 0$ $0.75 < \rho < \infty$

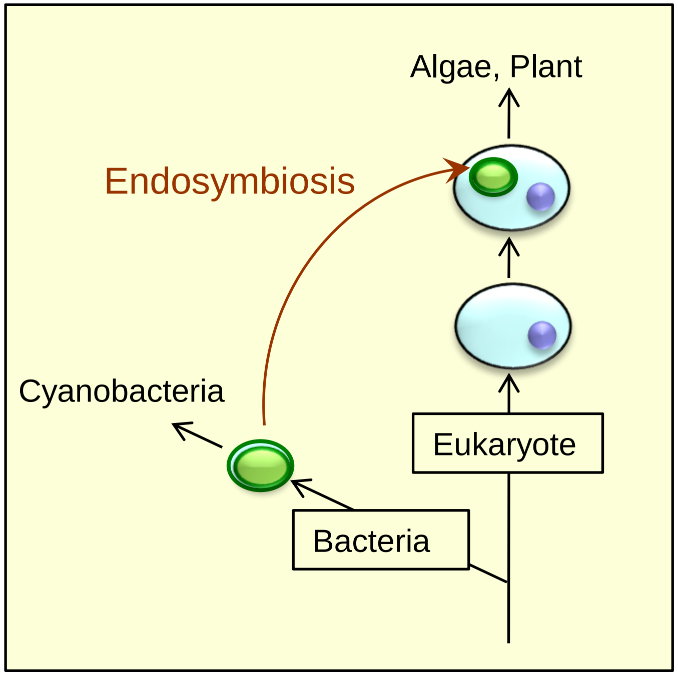
2) Non-degenerate condition (Hamaguchi, Harada, Shimanouchi,1975)

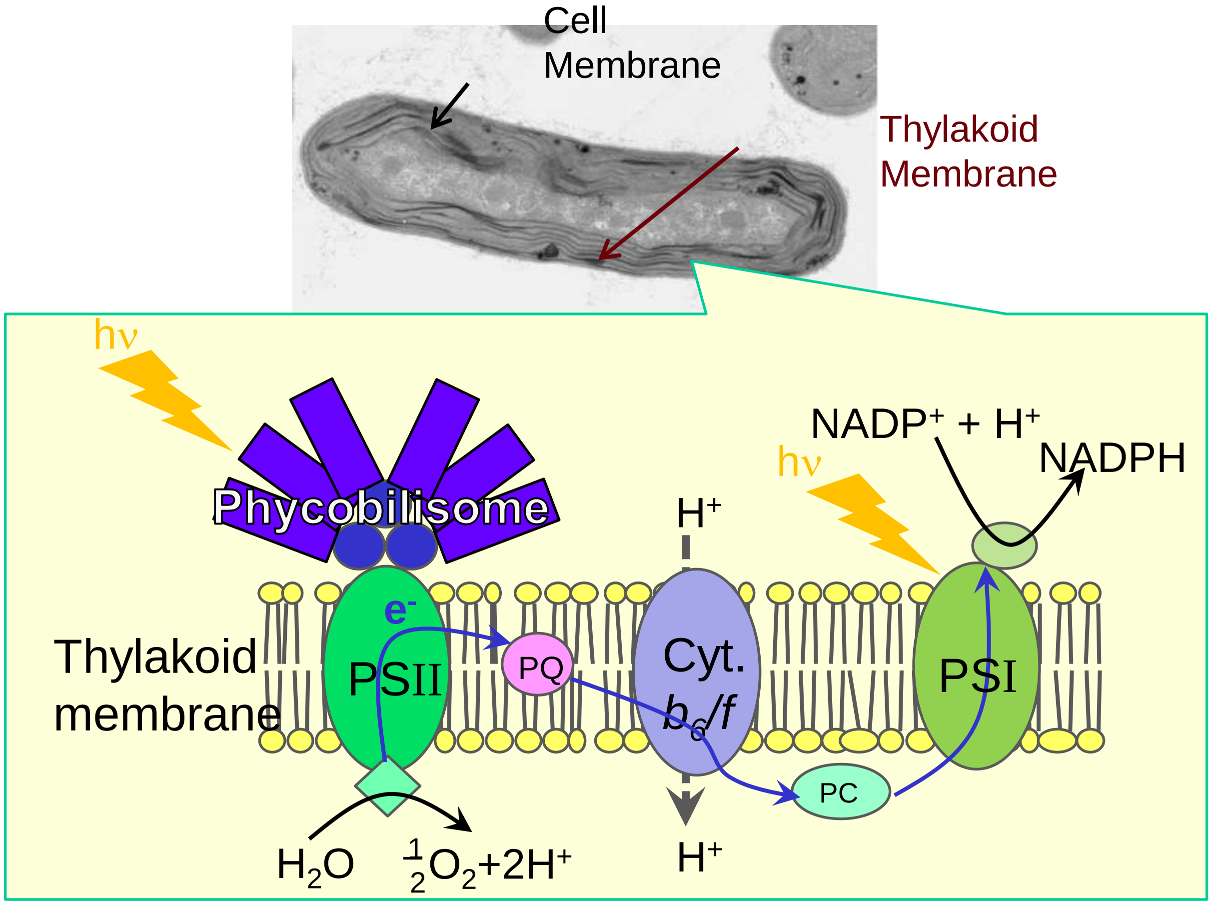
Totally symmetric modes : $G_0 \neq 0$, $G_a \neq 0$, $G_s = 0$ $0 < \rho < \infty$

Placzek’s prediction was proved after 40 years to establish firmly the theoretical basis of Raman spectroscopy.

Cyanobacteria

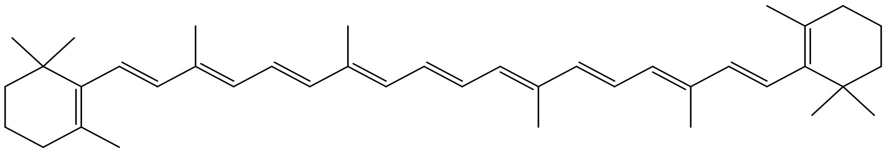
- Photosynthetic microorganism
- Origin of chloroplast
- Model organism for photosynthesis research



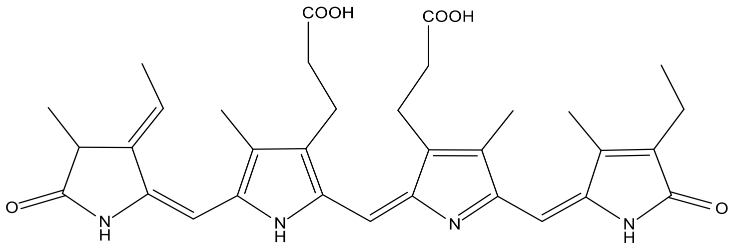


A. Herrero, E. Flores, *The Cyanobacteria: Molecular Biology, Genetics and Evolution.* Caister Academic Press (2008)

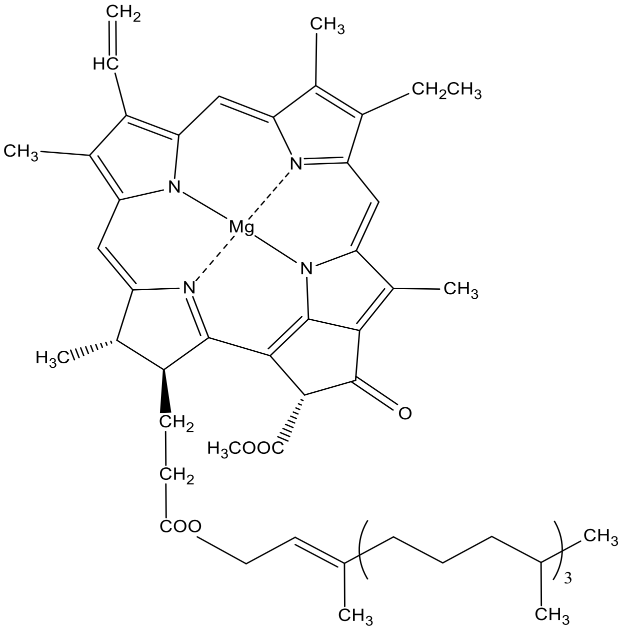
Pigments contained in cyanobacteria



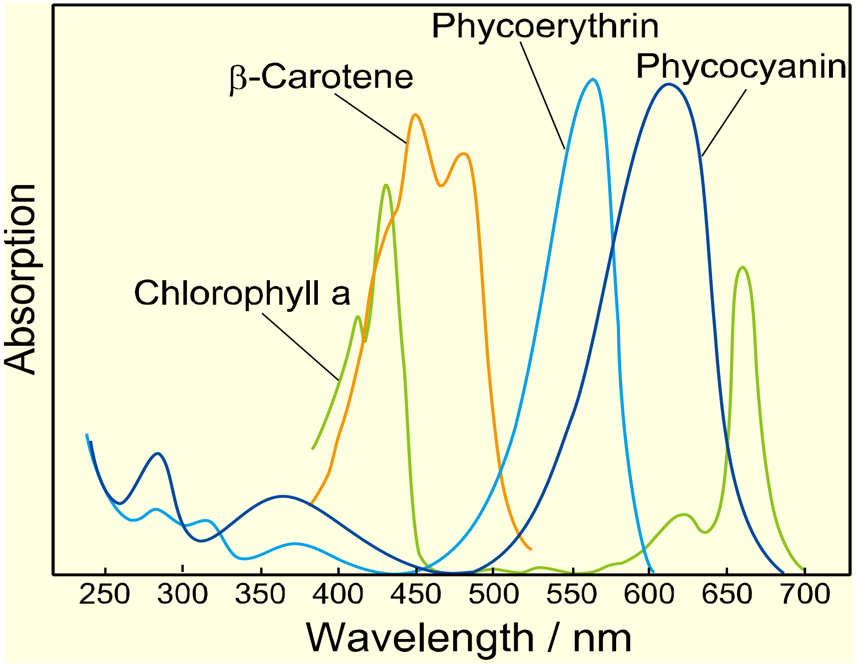
Carotenoid



Phycobilin



Chlorophyll a



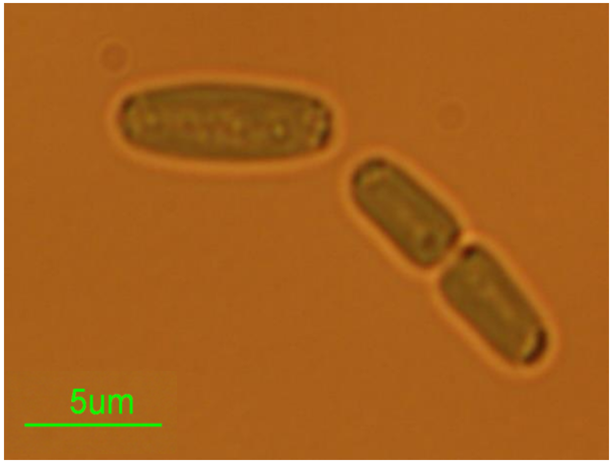
NIR Raman measurements of cyanobacteria

Thermosynechococcus elongatus

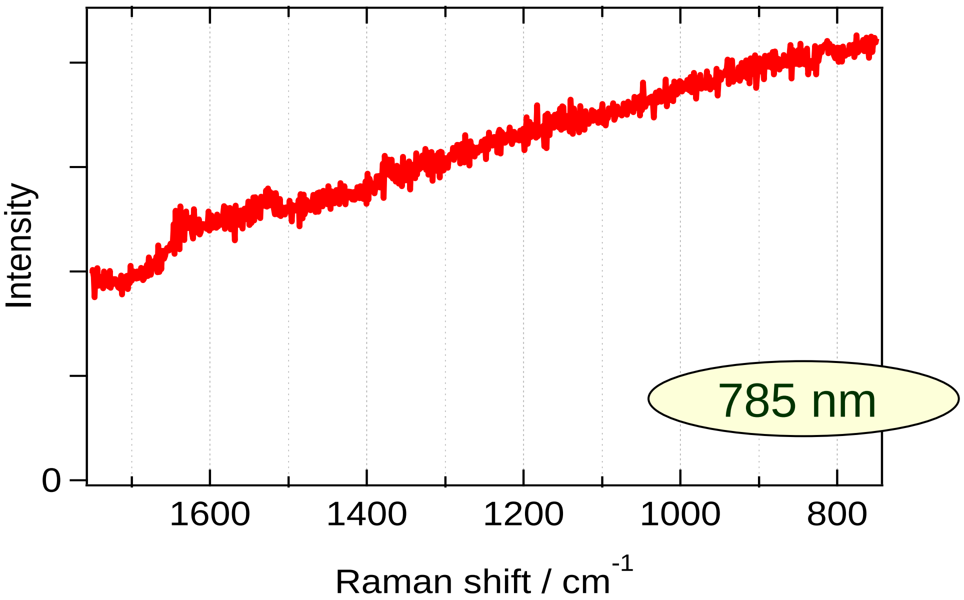
- Thermophilic cyanobacteria

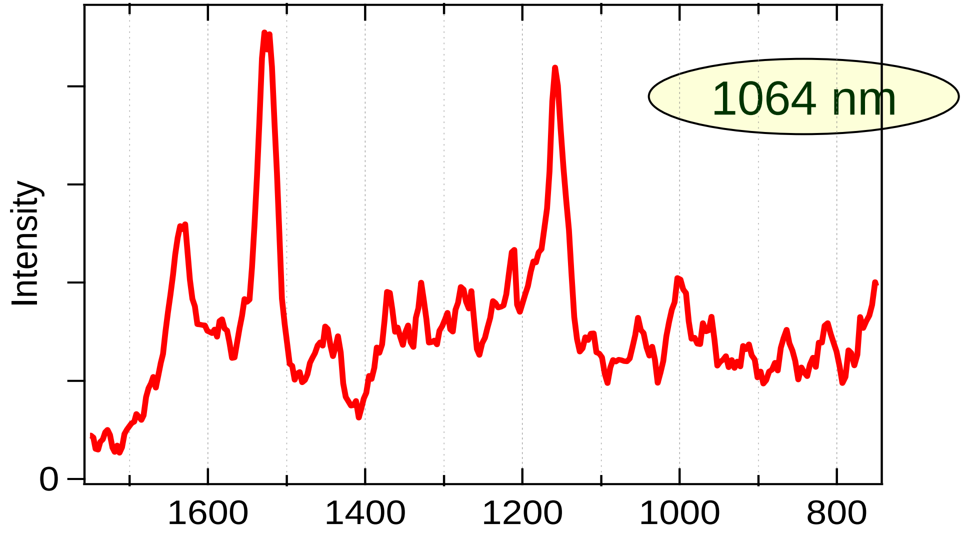
  (Unicellular rod-shaped)
- Well-established model organism

  -Entire genome sequenced

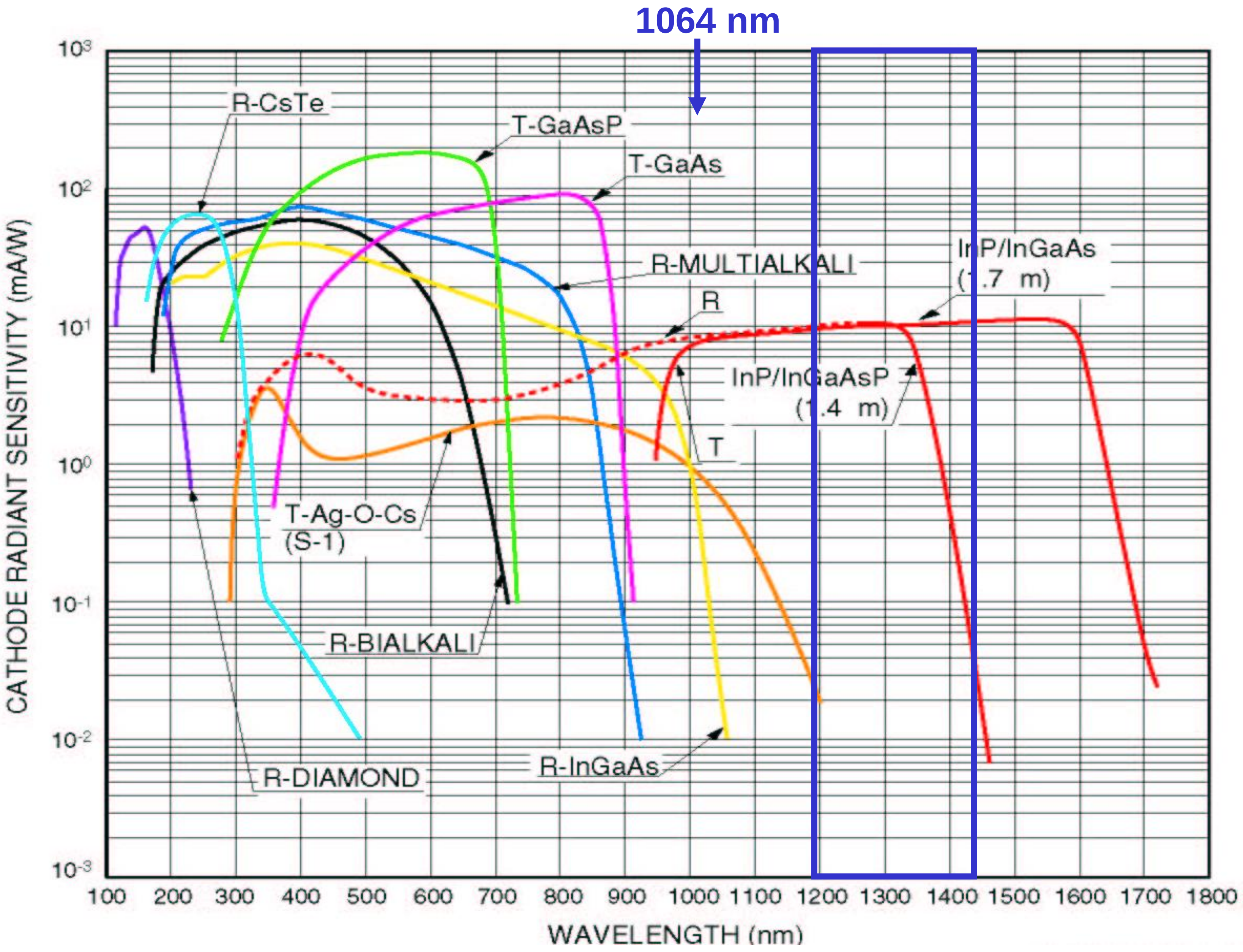


1 mW; 150 sec

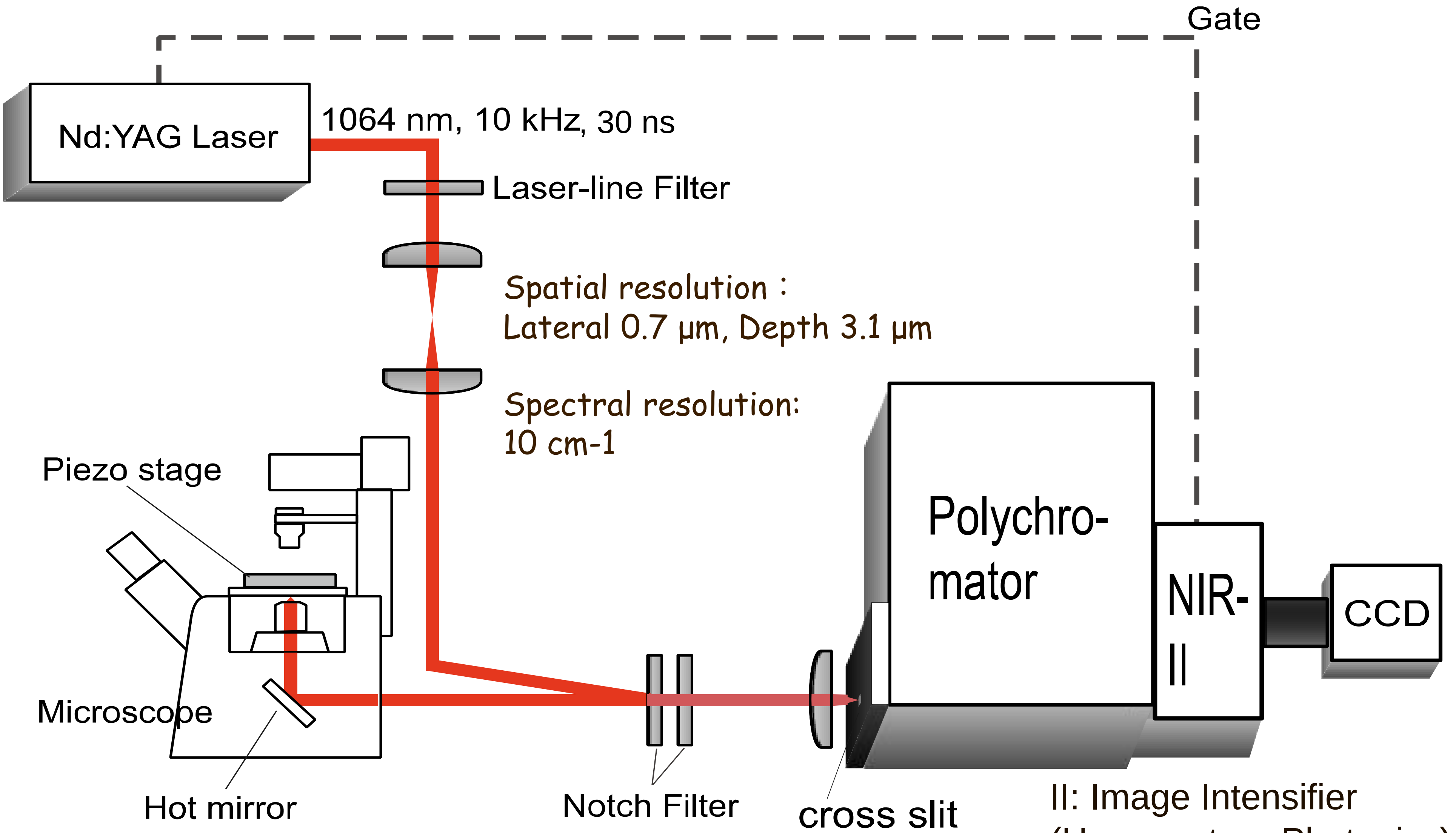




1064 nm Excited Raman Spectrpscopy with InP/InGaAsP Multichannel Detector



1064 nm Excited Multichannel Raman Microspectrometer

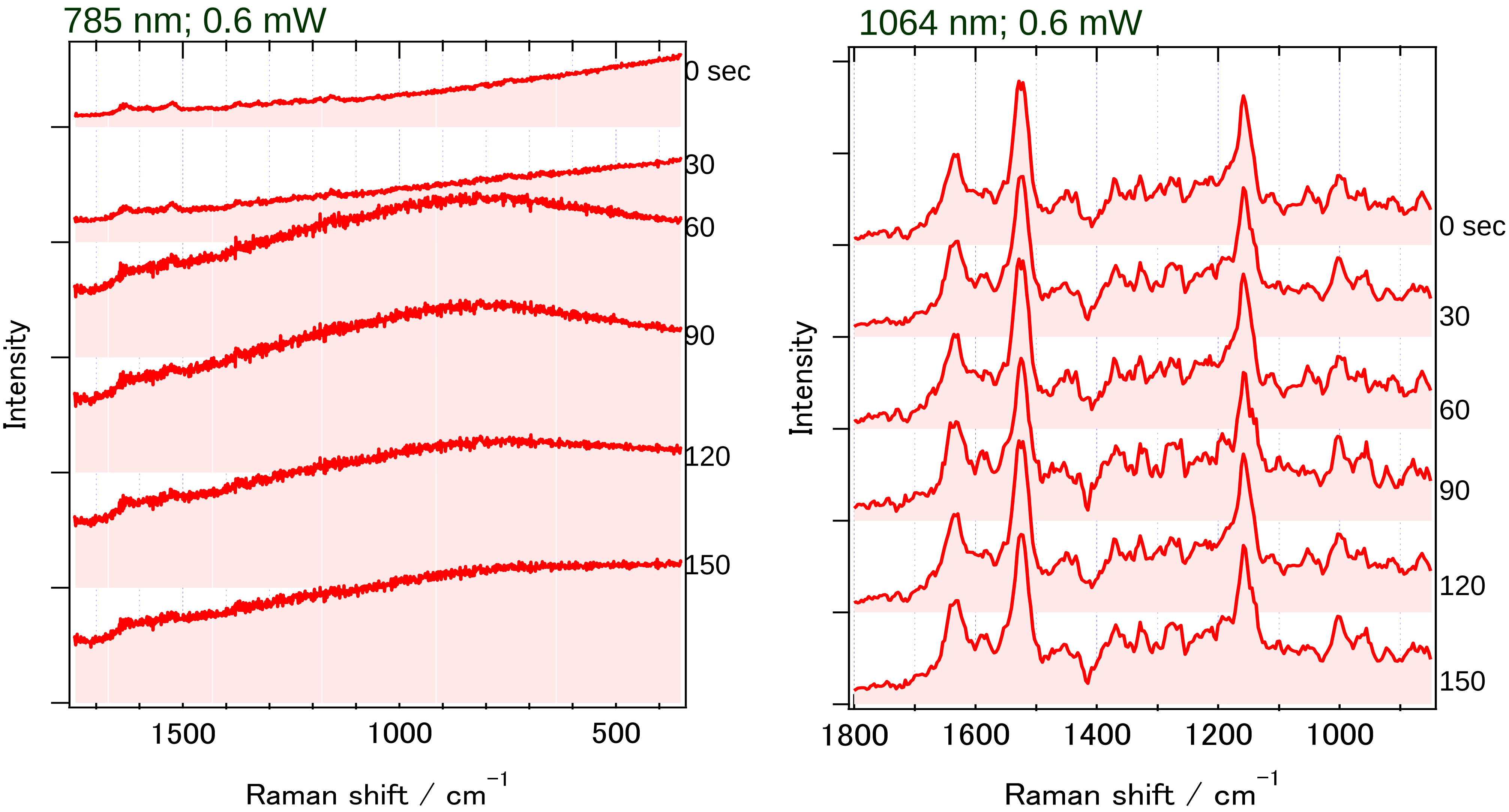


Spatial resolution :
Lateral 0.7 μm, Depth 3.1 μm

Spectral resolution:
10 cm-1

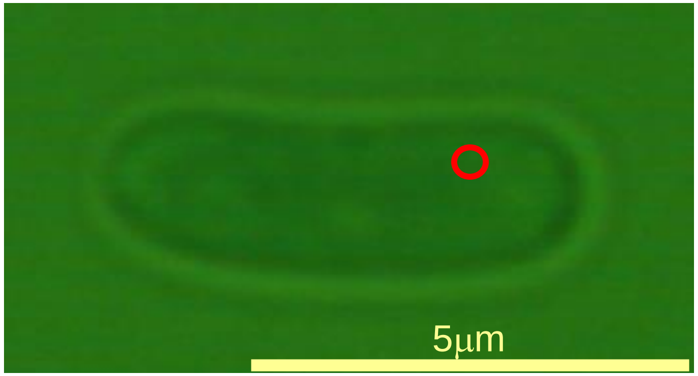
II: Image Intensifier
(Hamamatsu Photonics)

NIR Raman measurements of cyanobacteria



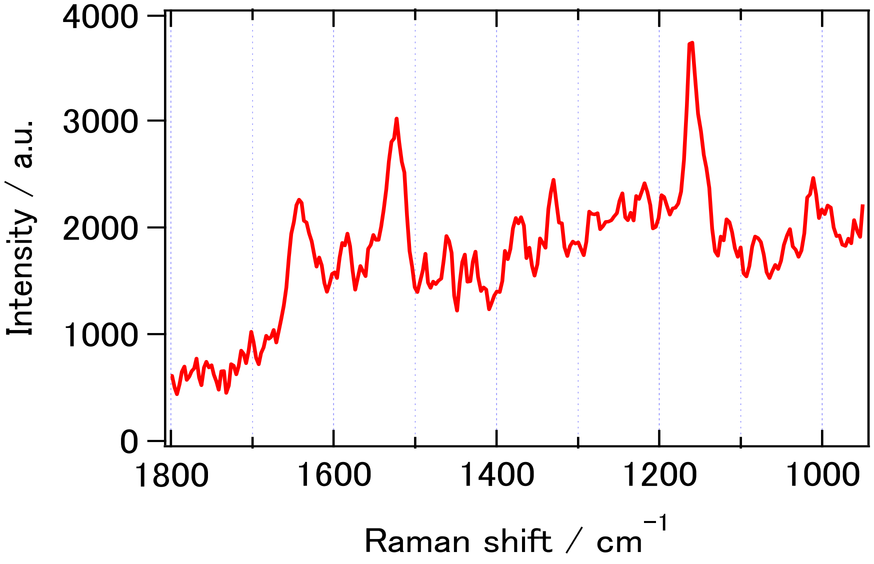
Deep near-infrared excitation is needed for avoiding the photodamage.

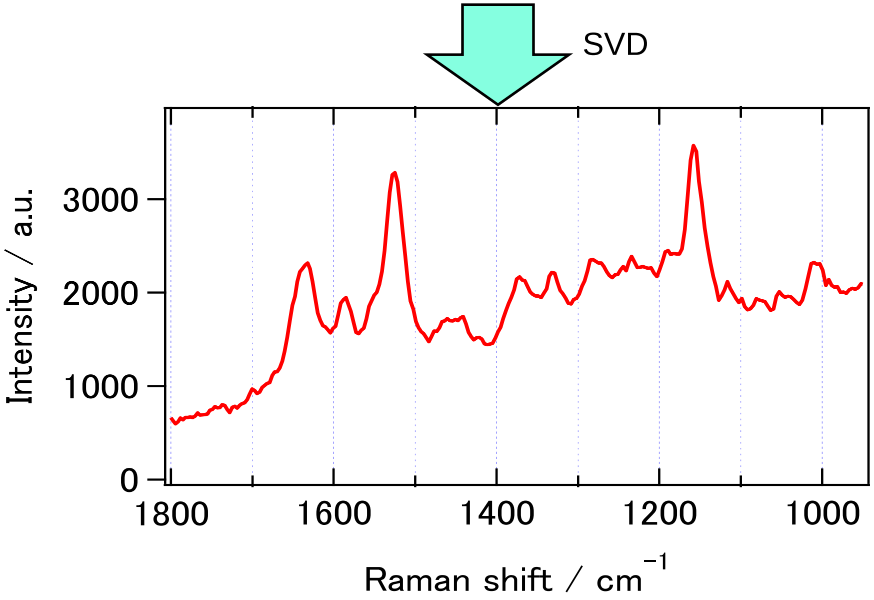
Space-resolved Measurements and SVD Noise Filtering



13 x 22 pix
(0.3μm interval)

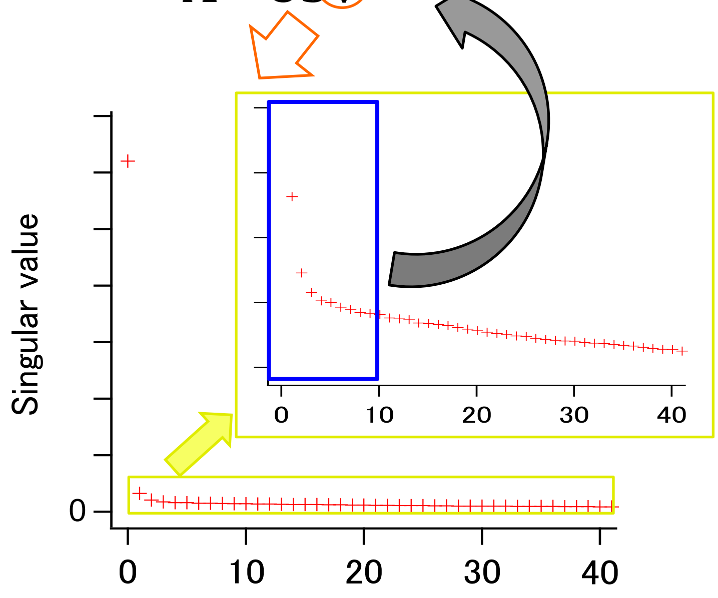
0.5 mW,
10 sec / pix





○ Singular value decomposition as noise filter1)

$$\mathbf{A} = \mathbf{USV}^{\mathbf{T}}$$



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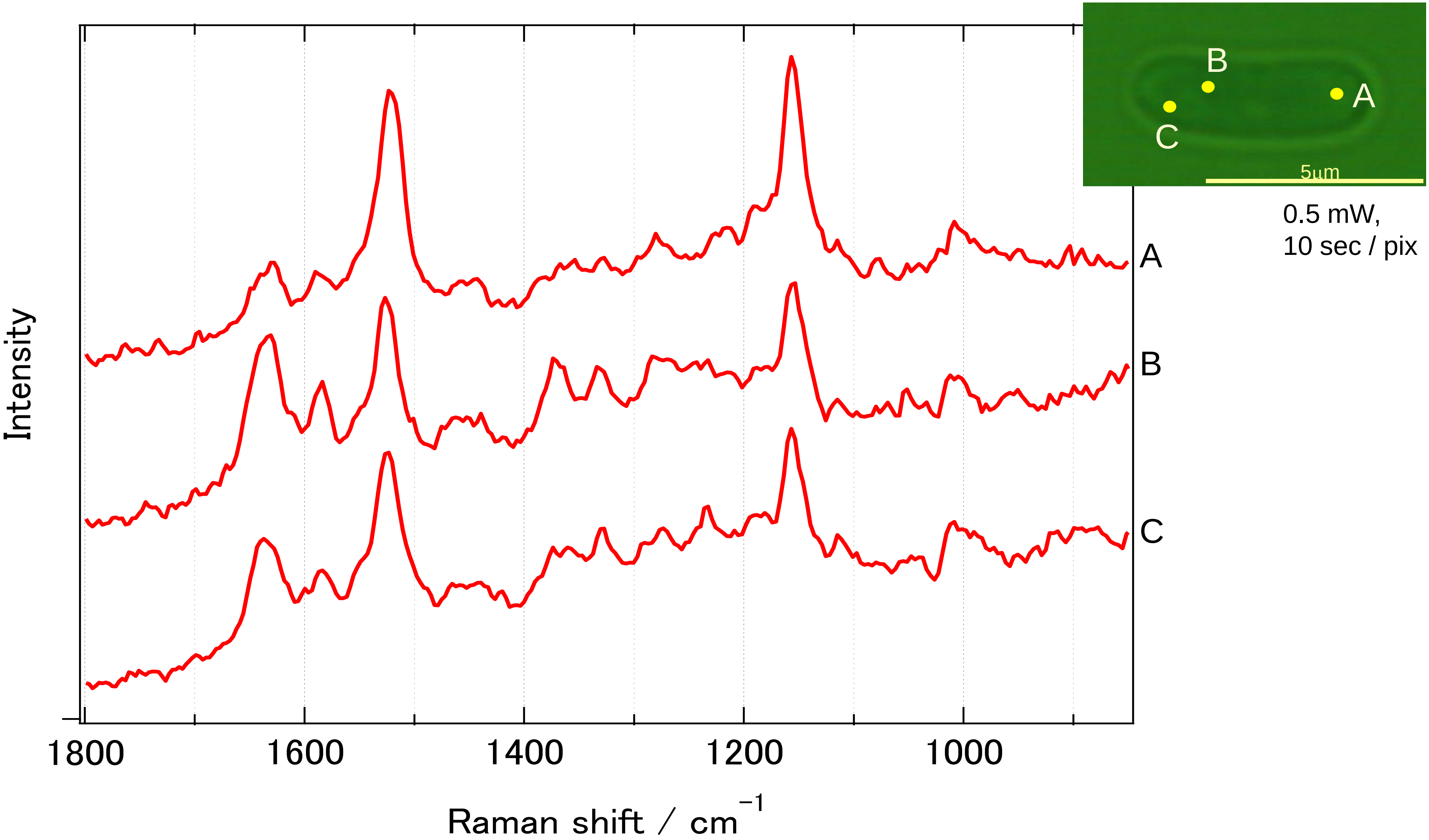
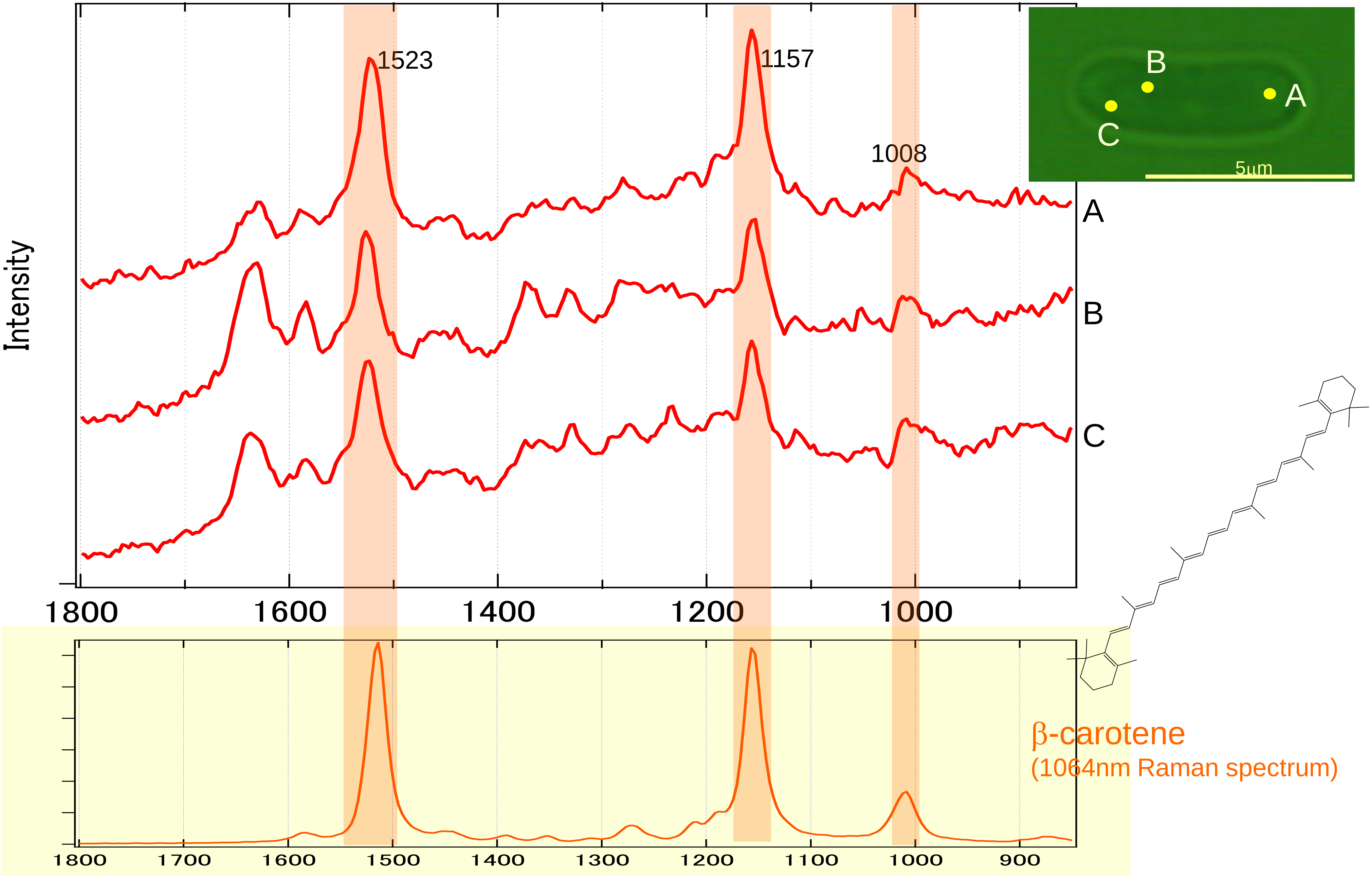
1) N. Uzunbajakava, C. Otto, et al. *Biophysical Journal* (2003) 3968-3981.

Space-resolved Raman Spectra within a Cell

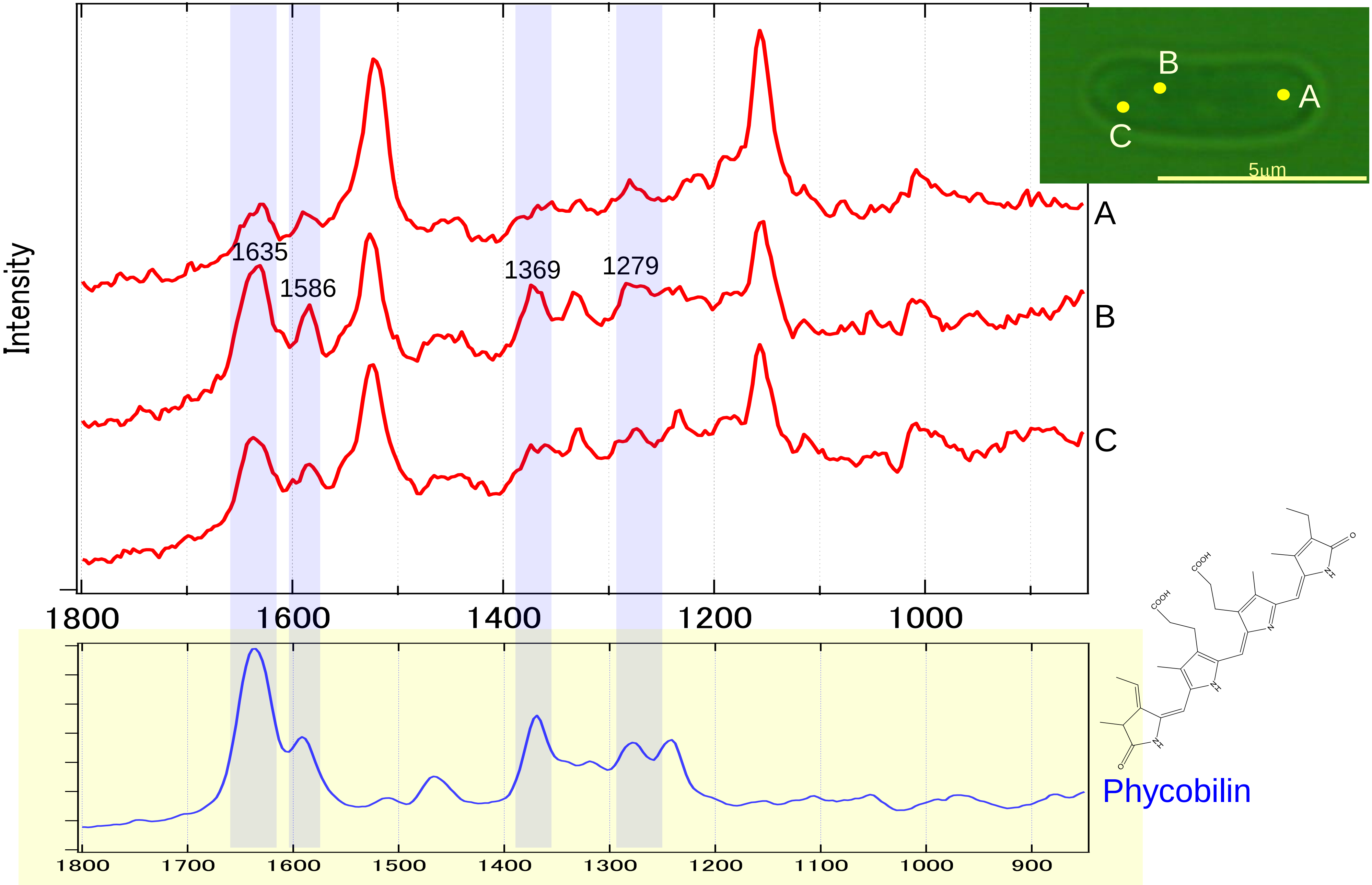


0.5 mW,
10 sec / pix

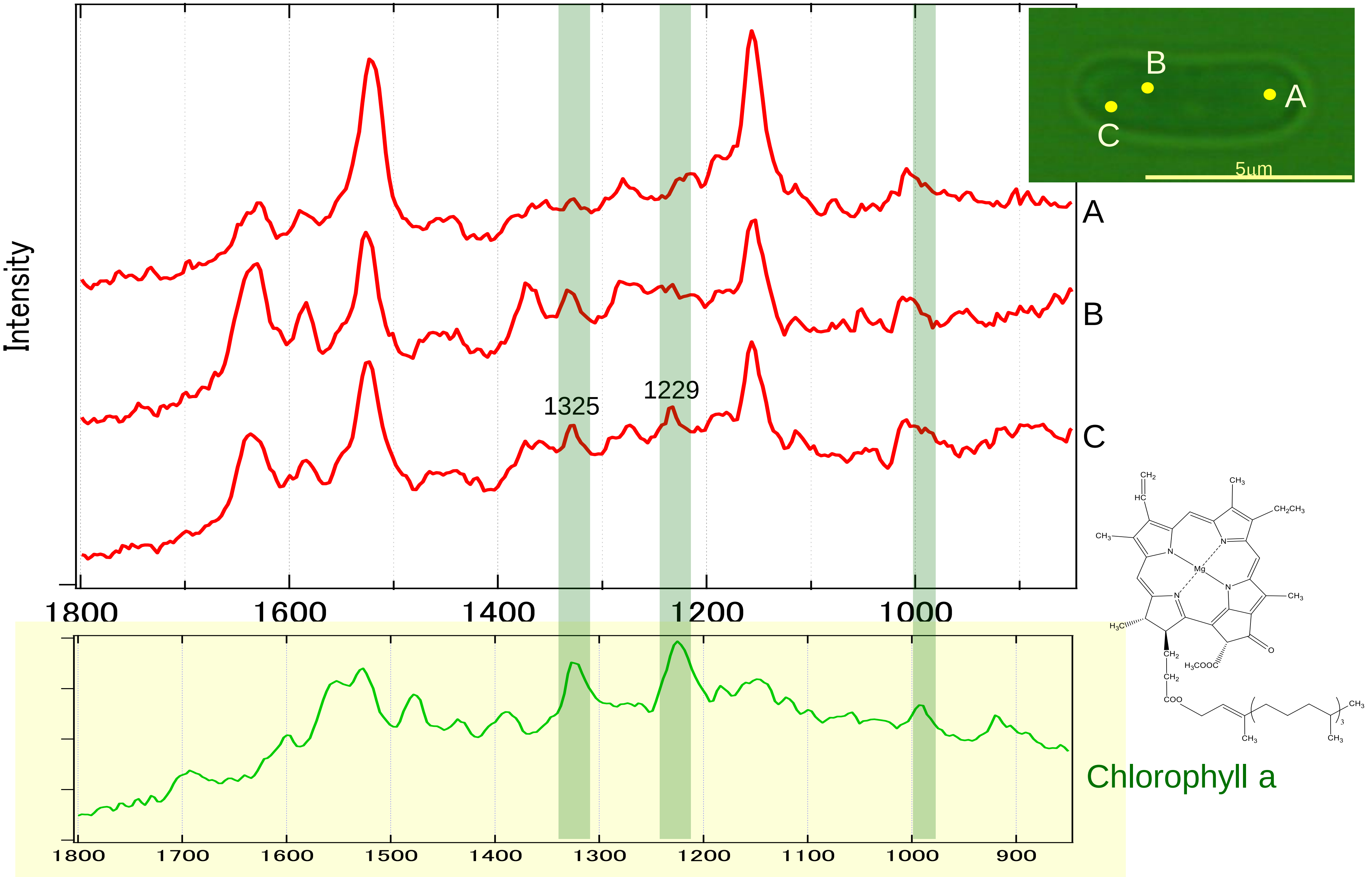
Band assignments



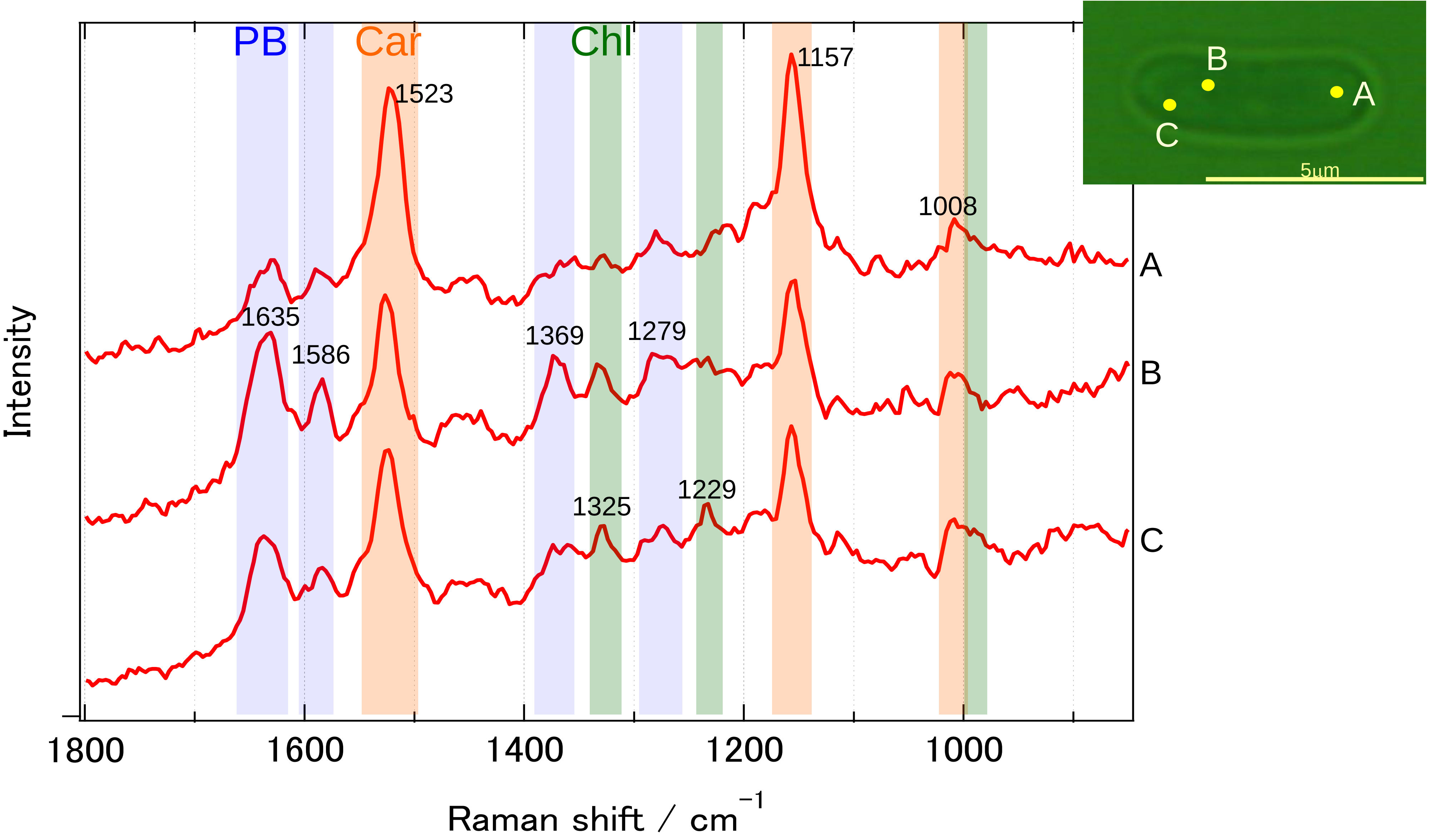
Band assignments



Band assignments



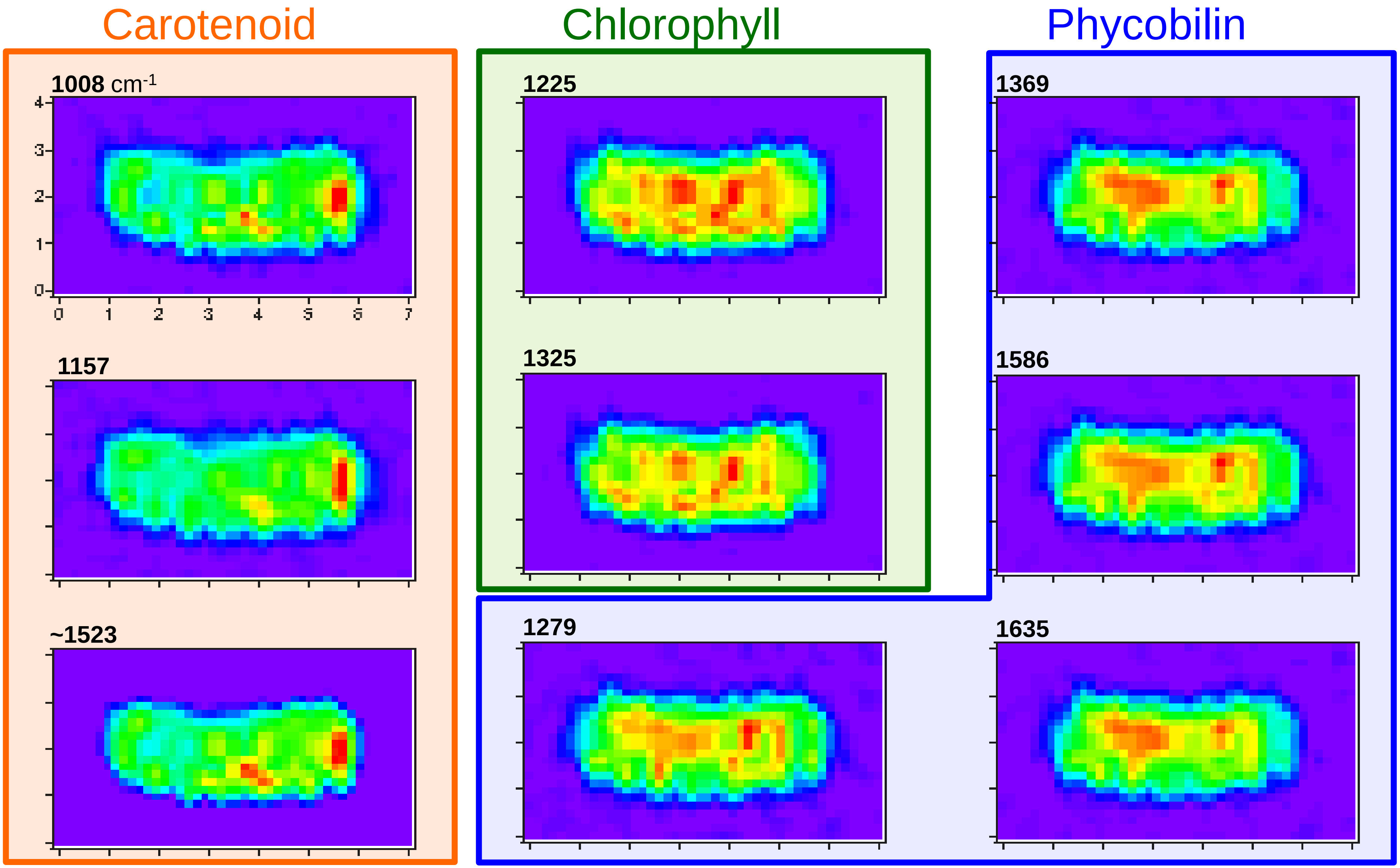
Band assignments



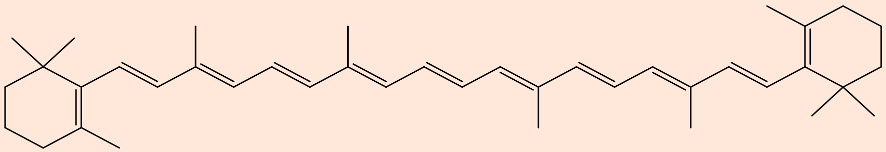
Almost all bands are assignable to the three photosynthetic pigments

⇐ Due to pre-resonance effect.

Raman mapping images

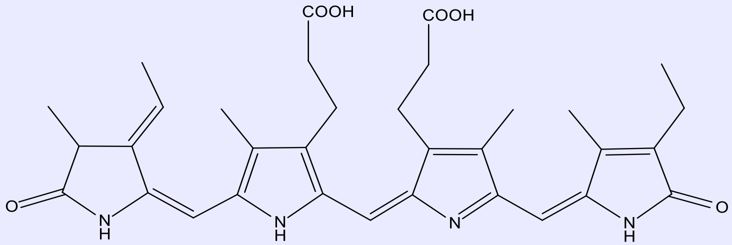


Functions of photosynthetic pigments



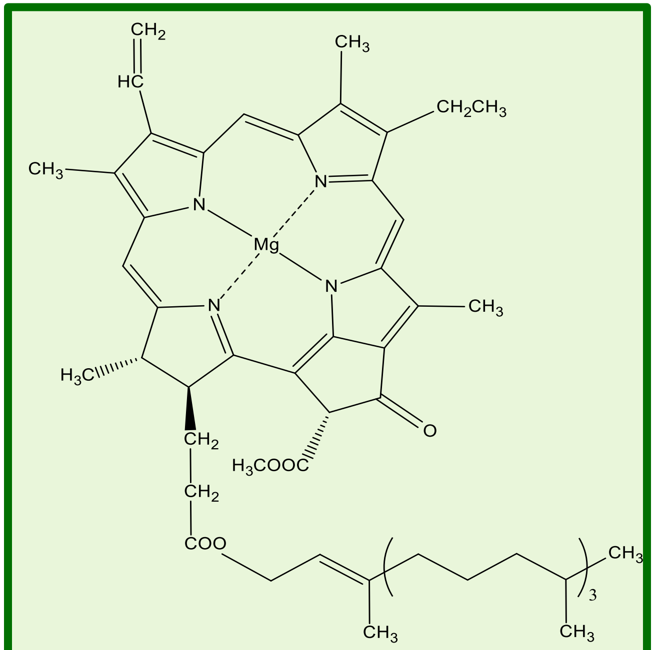
Carotenoid

Light harvesting
Antioxidant
Photoprotection



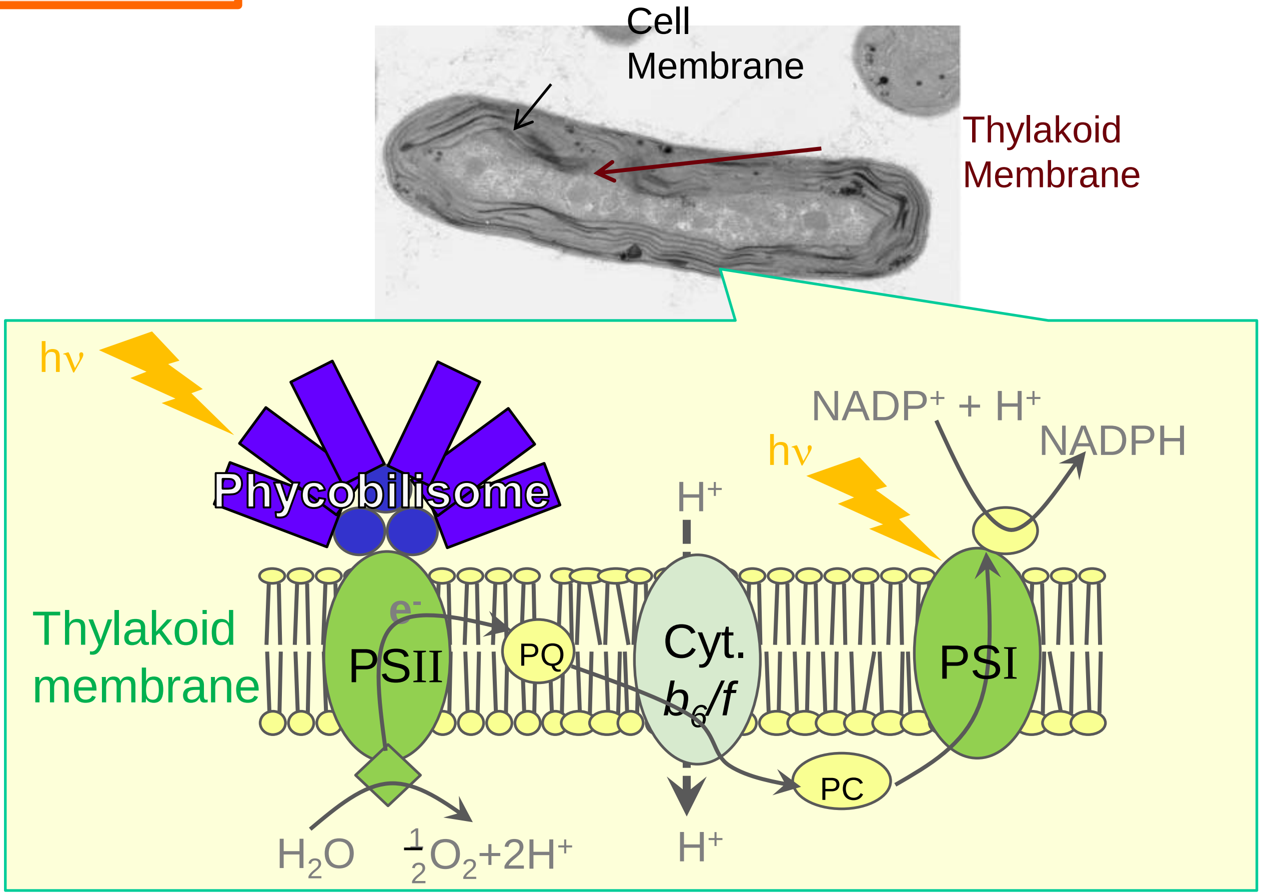
Phycobilin

Light harvesting

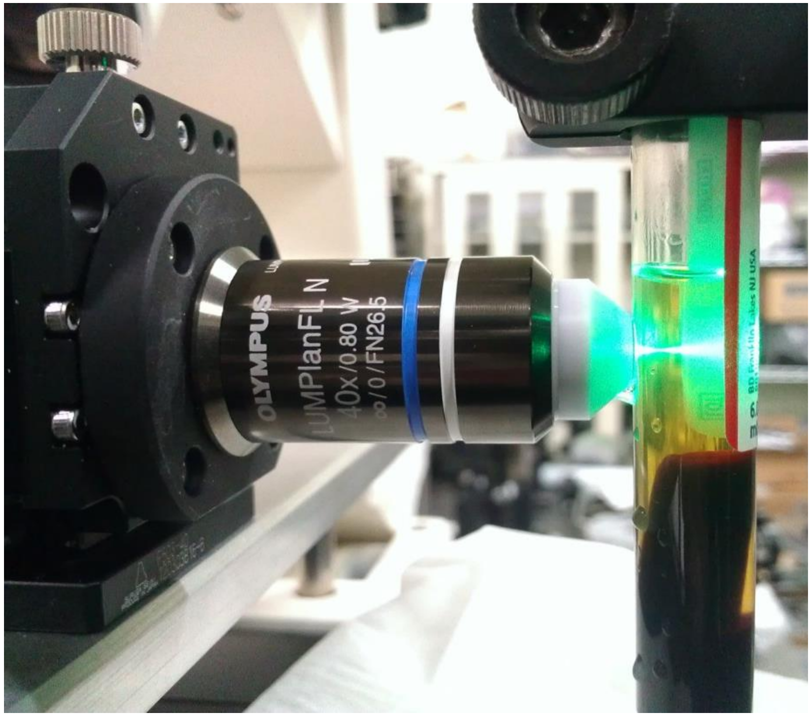


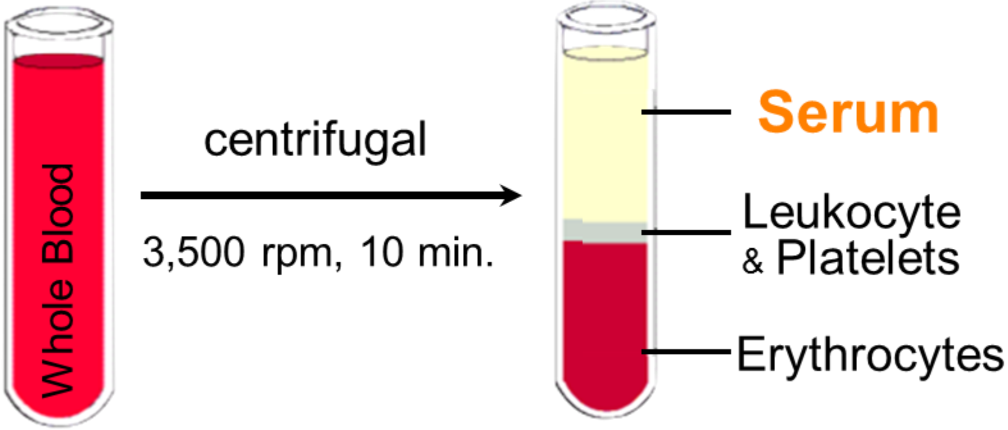
Chlorophyll a

Reaction center
Light harvesting



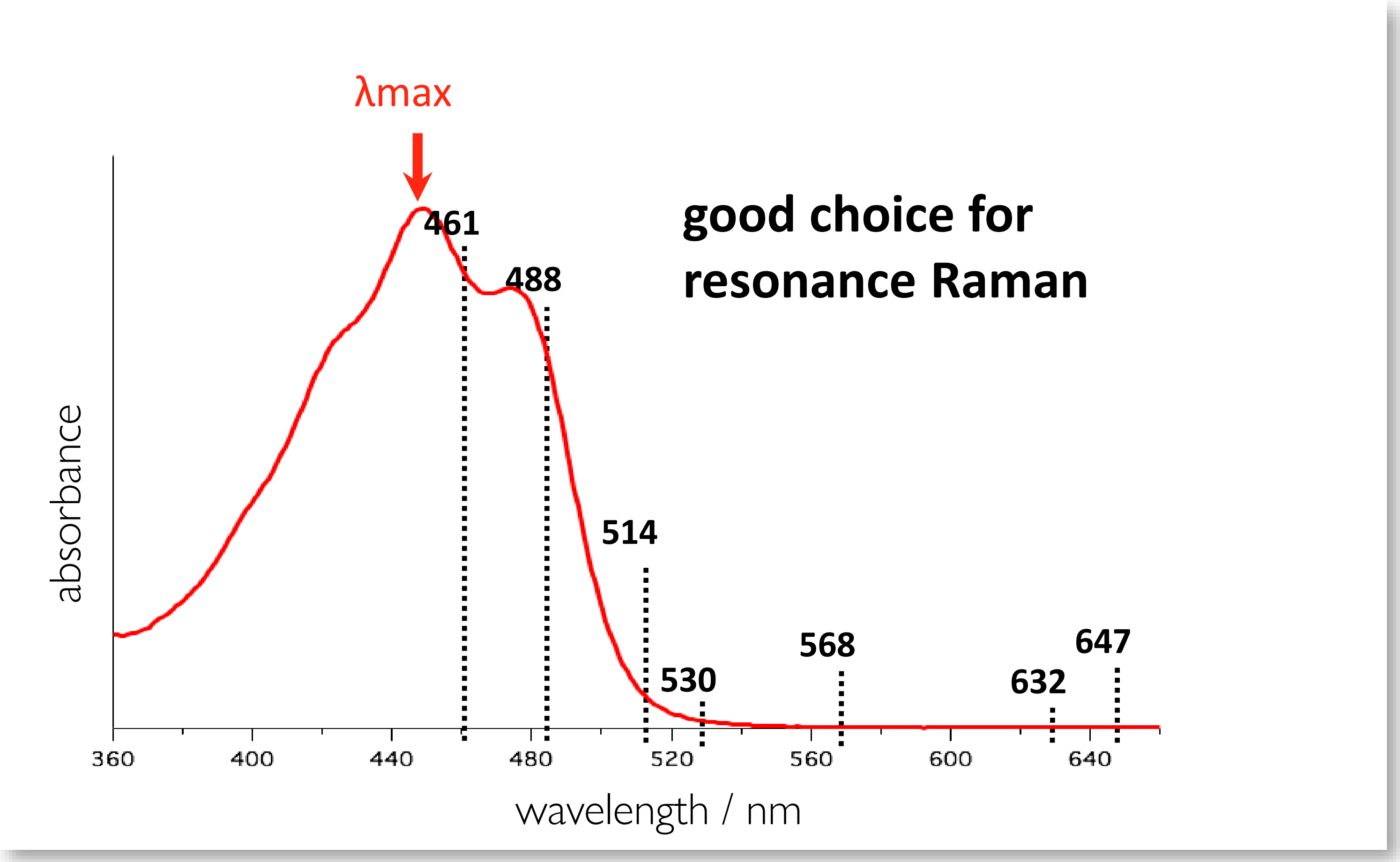
Resonance Raman Quantification of Carotenoids in Human Serum





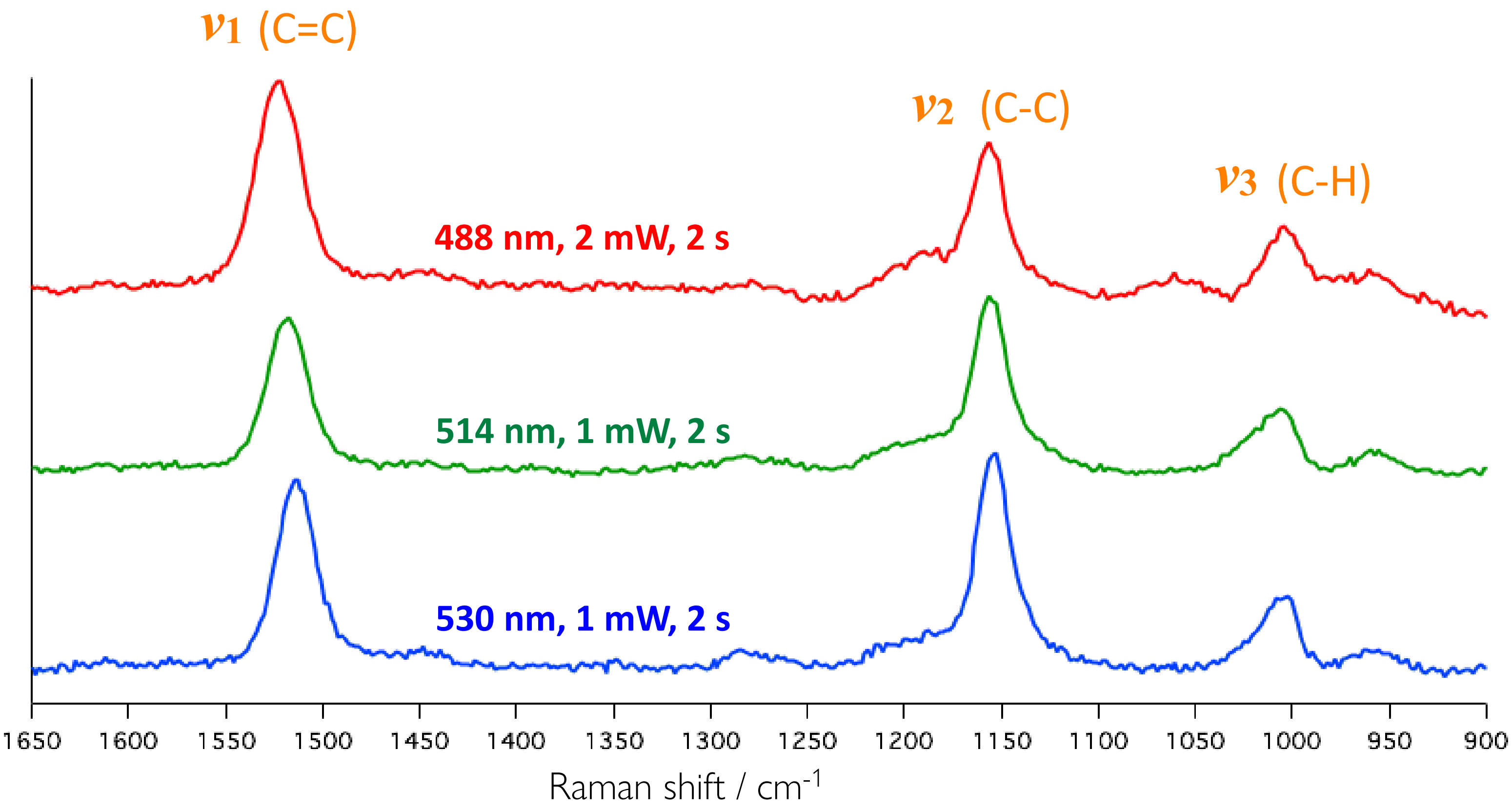
Absorption Spectra of ß-carotene / cyclohexane

(8 mg/L)

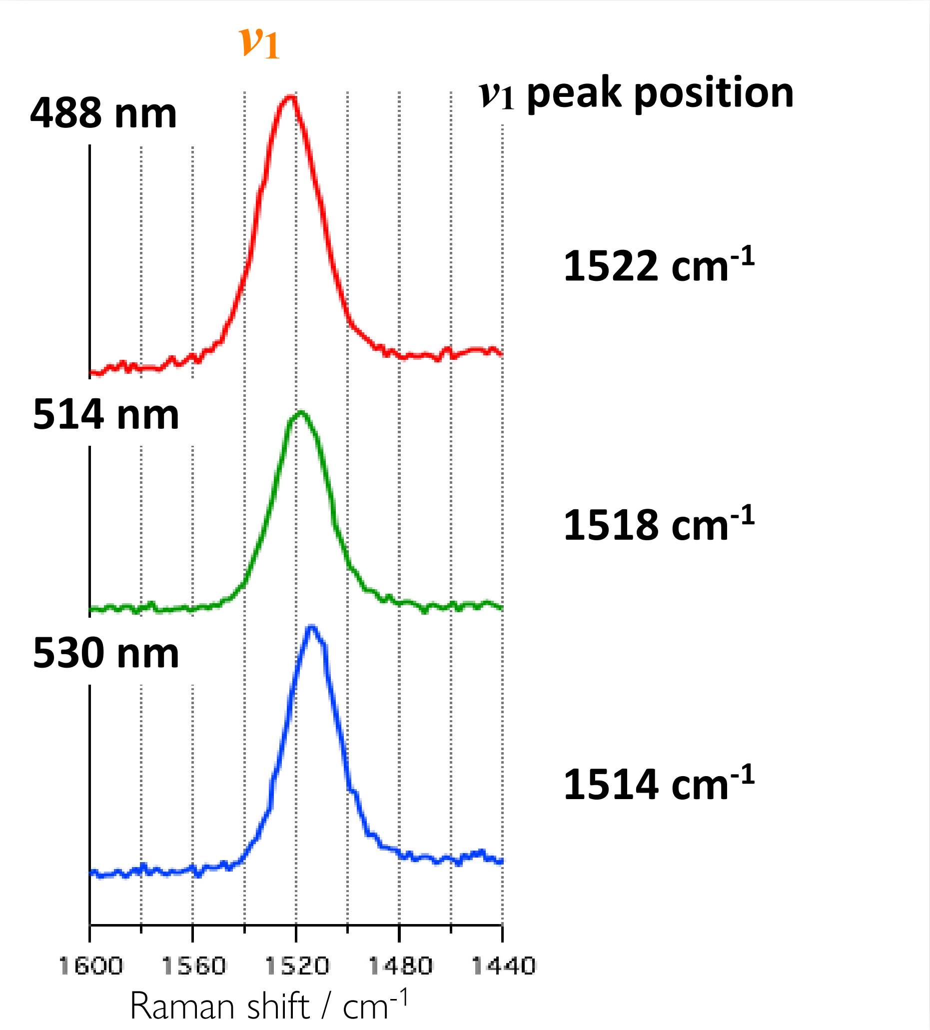


good choice for resonance Raman

Raman Spectra of Serum with Different Excitation Wavelength



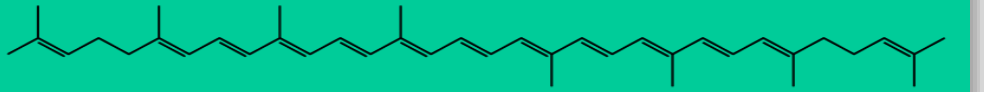
Raman Spectra of Serum with Different Excitation Wavelength



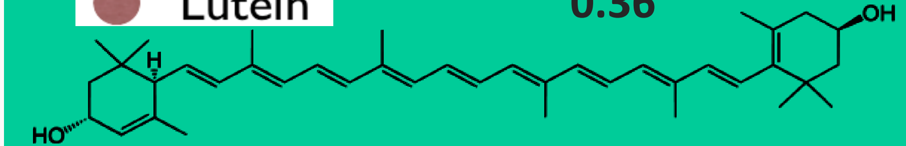
Characteristic of Serum Carotenoids

mean concentration
(µmol/L)

● Lycopene **0.40**

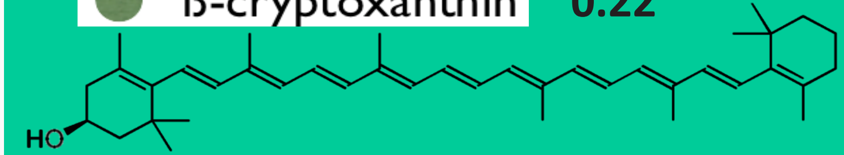


● Lutein **0.36**

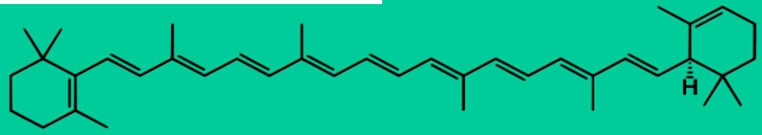


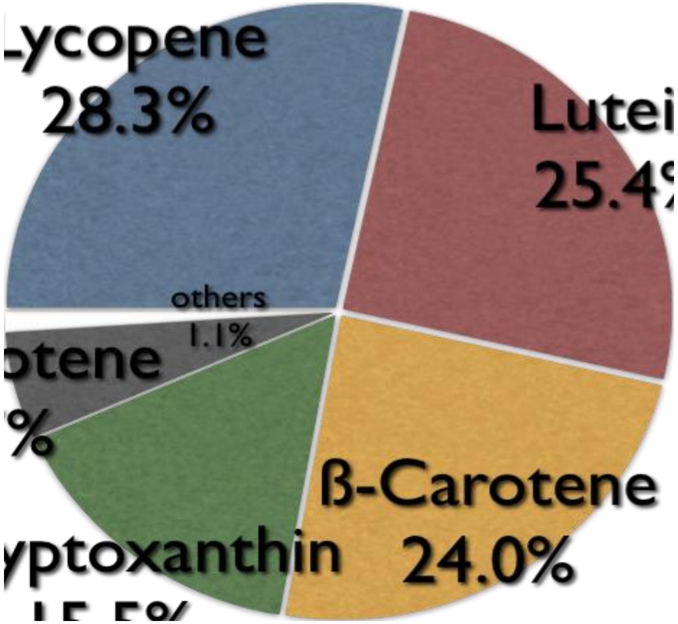
● ß-Carotene **0.34**

● ß-cryptoxanthin **0.22**



● α-Carotene **0.08**

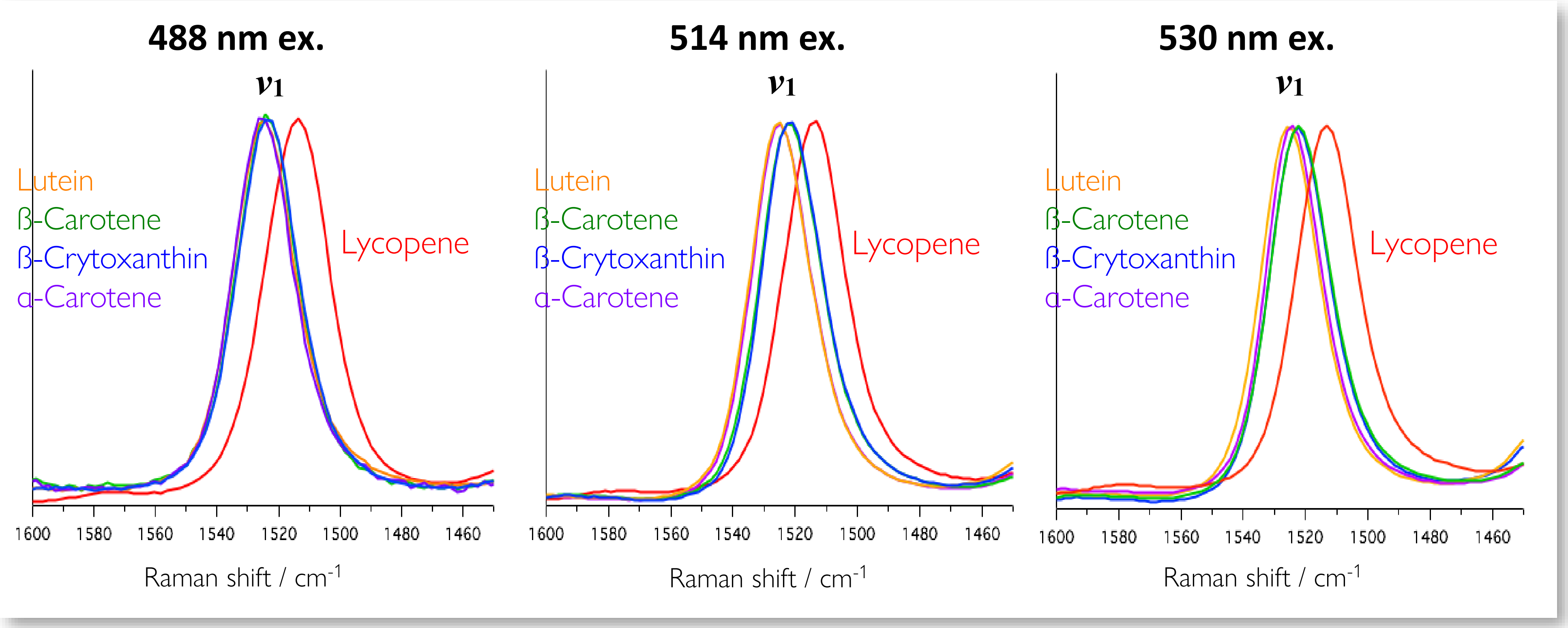




Proportion of individual carotenoids

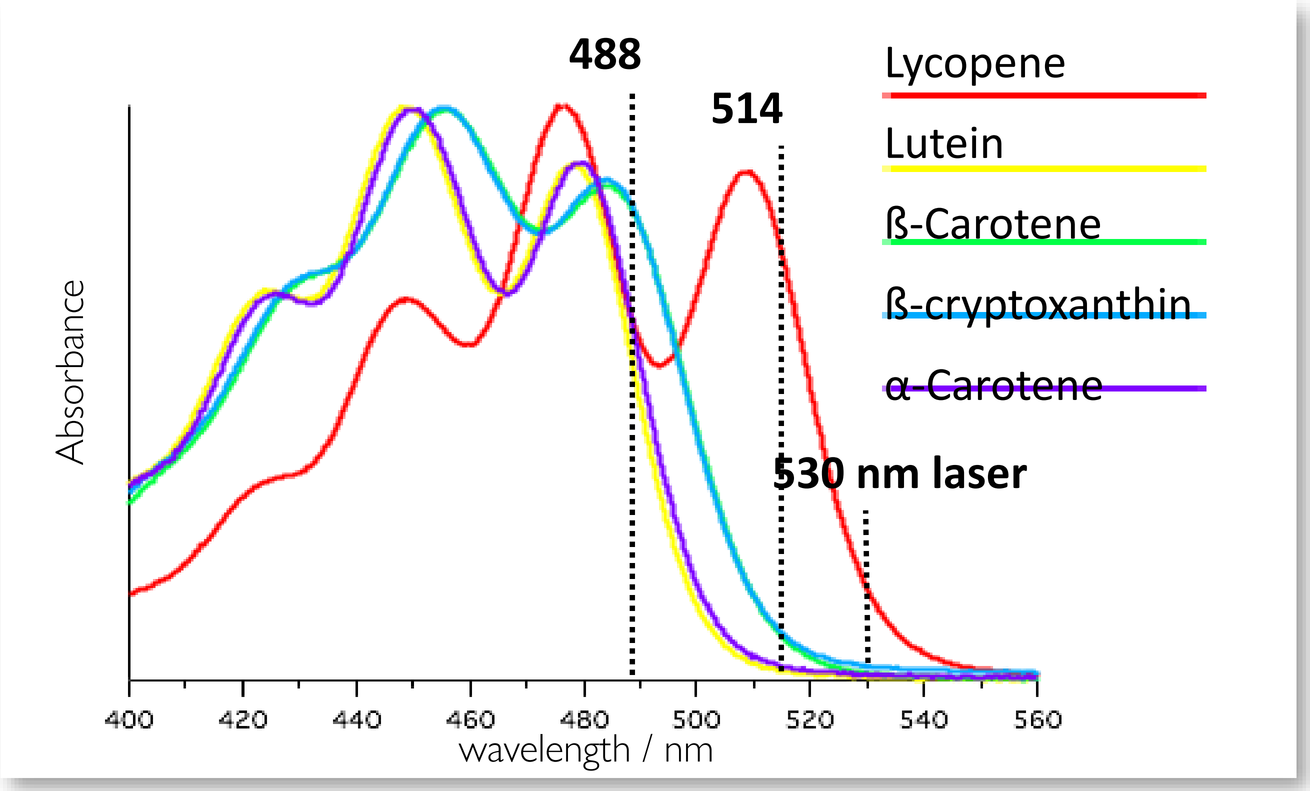
(E-Siong, 1999)

Raman Spectra of Carotenoids



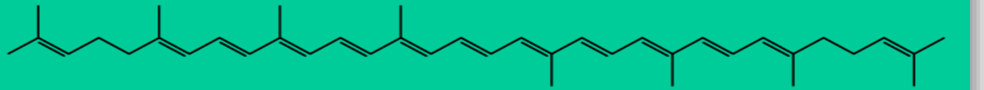
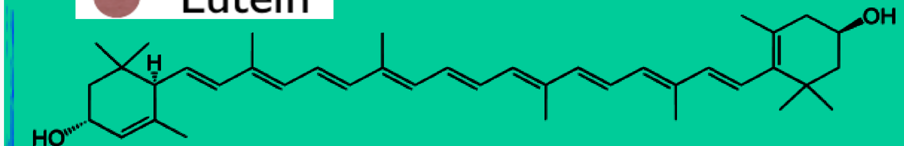
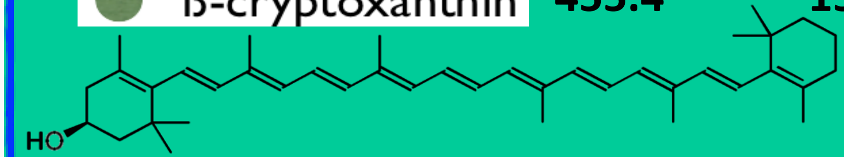
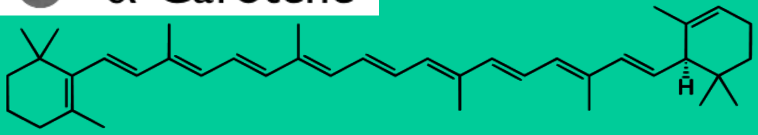
Lutein, ß-Carotene, ß-Crytoxanthin and α-Carotene have very similar v_1 peak position, apart from Lycopene.

Absorption Spectra of Carotenoids



Lutein, α-Carotene, ß-Crytoxanthin and ß-Carotene have very similar absorption characteristics, apart from Lycopene.

Two Groups of Carotenoids in Serum

	λmax (nm)	ν_1 (cm^{-1})	
● Lycopene	**476.5**	**1512.8**	
● Lutein	**449.0**	**1524.4**	
● ß-Carotene	**455.8**	**1523.7**	
● ß-cryptoxanthin	**455.4**	**1523.3**	
● α-Carotene	**450.1**	**1524.8**	

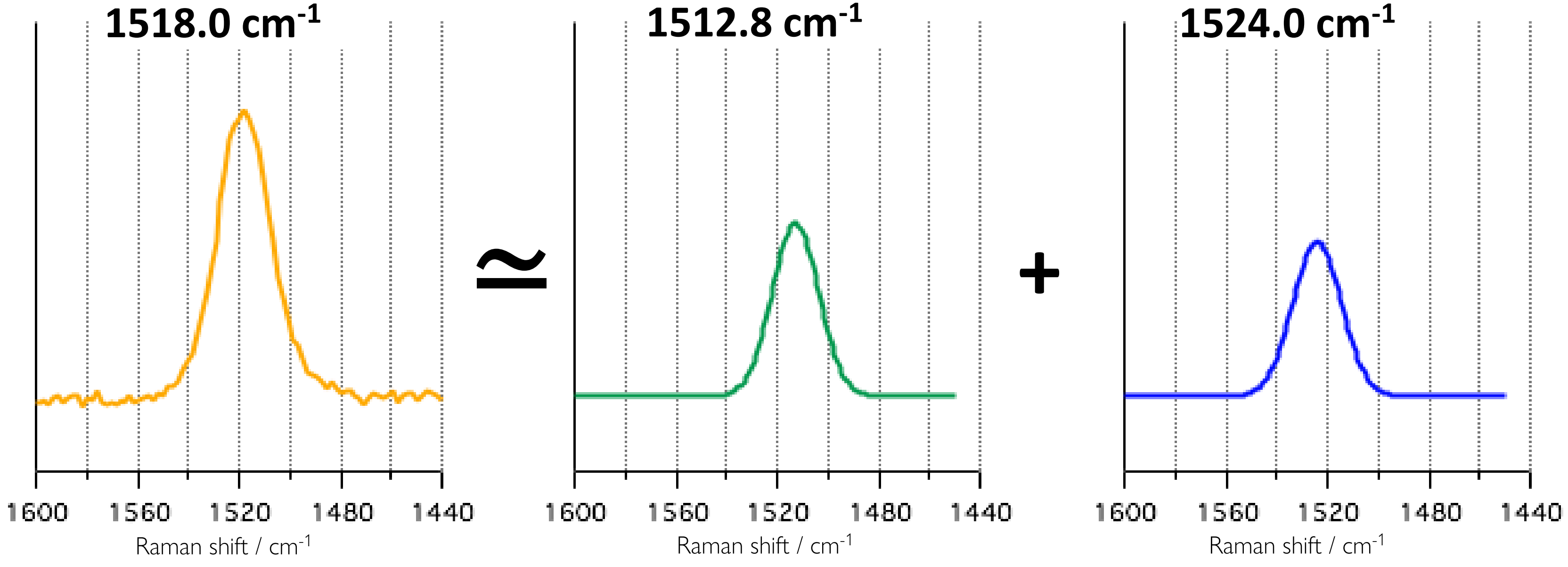
Component 1 : Lycopene

Component 2:

Represented by
ß-Carotene

(E-Siong, 1999)

Fitting the Observed Band with Two Components



Serum Carotenoids
ν_1

1518.0 $\mathrm{cm^{-1}}$

$\simeq$

Component 1
Lycopene

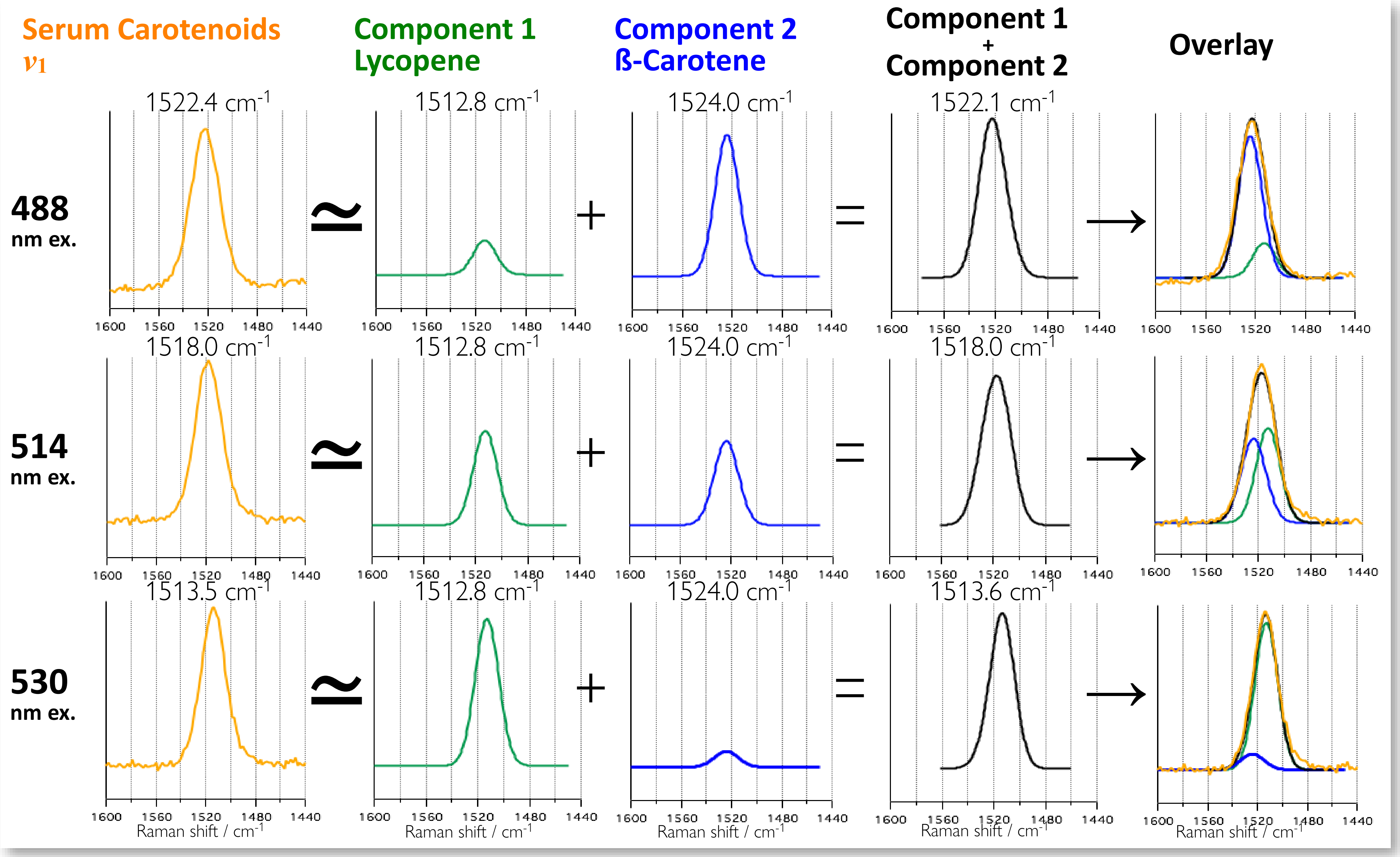
1512.8 $\mathrm{cm^{-1}}$

+

Component 2
ß-Carotene

1524.0 $\mathrm{cm^{-1}}$

A Global Analysis



Proportion of Components Amplitude after Global Analysis