Preparation and characterization of PVA-Chitosan-Gelatin composite doped with β-Carotenoids

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Abstract- In this study we prepared ternary blend film using poly vinyl alcohol-Chitosan-Gelatin composite (PCG). The prepared film was optimized using studies on melting temperatures, pH degradation, mechanical strength and electrical conductivity. Carotenoids were extracted from carrot using different solvents of concentrations. Carotenoids improve electrical conductivity due to delocalized π-electrons through conjugation. Hence various proportions of Carotenoids were doped to optimized PCG and electrical conductivity results were presented. It was found that electrical conductivity increases proportionally with Carotenoids concentration and saturates at a specific concentration. Thus this study will suggest the use of Carotenoids to enhance the electrical conductivity of biopolymers used in electronic applications.
Keywords: PVA, Chitosan, gelatin, Carotenoids, conductivity.

1. INTRODUCTION

The electrical conductivity of PVA / gelatin copolymer with different concentration of carrot carotene has been studied by Lofty [1]. The biodegradable property of PVA was studied by Hossam [2] and for PVA-gelatin composites were studied by Gamal [3]. Chitosan has bacteriostatic effects and so enhance wound healing according to Nazar [4]. Thermal, structure and surface morphology for Chitosan-PVA-gelatin composite was studied by Cheng [5]. Lihong [6] prepared Chitosan-PVA-gelatin composite using gamma irradiation and studied its application for wound healing. Leatherman [7] found that color in plants and micro organism is due to absorption of light by carotene and hence the carotenes are readily oxidized electrochemically implying that they enhance electrical conductivity.

2. MATERIALS & METHODS

2.1 Materials

Poly vinyl Alcohol (PVA) was purchased from Meru Chem pvt. Ltd. Chitosan (95% de-acetylated) was purchased from Panvo Organics pvt. Ltd. Gelatin from M. Pharmaceuticals, Glacial acetic acid from Aditya commercial corporation, Ethanol 95% and from sugarcane Industries and petroleum ether from Hi-Chem.

2.2 Methods

2.2.1 Preparation of PVA solution (A)

100 ml of distilled water is taken in to a 500 ml beaker and heated until water gets boiled. Once water bubbles comes, then stop heating water and to this 2.5 mg of PVA is added and stirred until clear solution of PVA obtained. This 100 ml of PVA solution is denoted as “A”.

2.2.2 Preparation of Chitosan solution (B)

100 ml of distilled water is taken in to a 500 ml beaker and heated along with 2.0 gm of Chitosan. The solution was stirred with pellet magnetically at a speed of 900 rpm for 15-20 minutes until Chitosan dissolves completely. Then 6 ml of 1 M glacial Acetic acid is added and the solution is allowed to cooled for 10 minutes. Thus a 2% Chitosan solution was prepared and is denoted as “B”.

2.2.3 Preparation of Binary Blend (AB)

100 ml of above prepared PVA solution is added to 100 ml of above prepared Chitosan (CHI) solution in a 500 ml beaker and heated the resultant solution until a homogeneous solution is formed. This 200 ml solution is denoted as “AB”.

2.2.4 Preparation of Gelatin solution (C)

5 gm of gelatin was added to 100 ml of boiled distilled water and stirred the solution at a speed of 500 rpm until a clear solution is formed. This 5% gelatin solution is denoted as “C”.

2.2.5 Preparation of Optimized Ternary Blend

200 ml of AB and 100 ml of C were added in different proportions as shown in below table and the samples were labeled as I, II, III, IV, V and VI.

Table.1. Blending ration of AB and C films

Sample	Solution-AB (ml)	Solution-C (ml)
I	2	13
II	4	11
III	6	9
IV	8	7
V	10	5
VI	12	3

Above prepared 6 samples were poured into different glass Petri dishes and kept them in an oven at 40^oC for 24 hr to form films and are denoted as Sample-1, Sample-2, Sample-3, Sample-4, Sample-5, Sample-6 respectively.

2.2.6 Extraction of Carotenoids (Ethanol)
200 gm carrot cut in to small pieces and dried for about 10 min in an oven at 60^oC for about 30 minutes. Then take in to a beaker and 250 ml of 90% ethanol is added. The mixture was stirred thoroughly for about 10 minutes and allowed to rest for 24 hours. Ethanol was filtered and allowed to evaporate up to 200 ml and equal amount of petroleum ether is added to obtain a homogenous mixture. Adding 3 ml distilled water to the solution causes the phases to separate in to yellow carotene rich phase and translucent / green phase. Then carotene rich phase is isolated from the solution.

2.2.7 Carotenoids Extraction (Propan-2-ol)
350 gm of carrot sliced, grounded and mixed with 500 ml propanol. After 24 hr equal amount of pet ether is added and carotenoid extract is collected and stored.

2.3 Optimization of ABC composite
The above six samples were optimized for melting point, Tensile strength, pH degradation and DC/AC electrical conductivity.

2.3.1 Melting point Analysis
The above six films were individually tested for melting point by keeping them in a vacuum oven and temperature is slowly increased.

2.3.2 Tensile strength
The samples ABC were characterized for their Tensile strength using INSTRON 1405 at a speed of 5 mm/min. Two dumbbell shaped specimens of 5 mm wide and 10 mm length were punched out using a die. Mechanical properties such as Tensile strength (MPa) and elongation (%) were measured.

2.3.3 pH degradation
200 ml of 10X PBS buffer with pH 7.4 was prepared using components listed in table.3.

Table.3 Preparation of 10X PBS buffer

Chemical	Amount (gm)
Na_2HPO_4	0.284
KH_2PO_4	0.0488
NaCl	1.6
KCl	0.04

Individual films were placed in glass Petri dishes and equal volume of 10x PBS buffer of pH 7.4 was added to them. The films were checked for partial and total degradation and noted time was tabulated.

2.3.4 Preparation of conductive films
To each ternary solution (15 ml), different amounts (0-5%) of carotene extract is added to get six samples. These samples were poured into petri dishes and placed in an oven at 40^oC for 24 hr to dry.

2.3.5 DC Resistance
Electrical DC resistance of the films AB & ABC were determined by using "Mastech MAS830L Digital Pocket Multimeter".

2.3.6 AC Resistance
AC resistance of the films AB & ABC were determined by using "Mastech MAS830L Digital Pocket Multimeter". The electrical circuit to determine ac resistance of the films is given below fig.1. Sample is placed between copper plates to give good electrical contact.

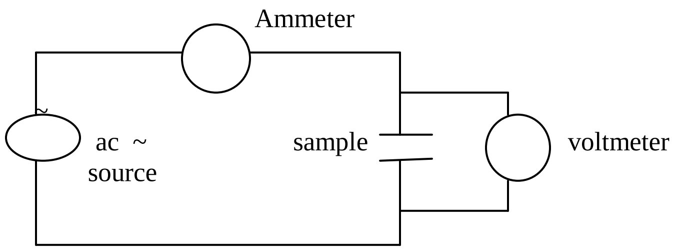

Fig.1. Circuit diagram to measure ac resistance

2.3.7 FTIR Analysis
FTIR spectroscopy for samples is analyzed using Nicolet 170 SX, Thermo Fisher Scientific Inc, USA in the range of 400-4000 cm-1. One mg of dry sample was mixed with 100 mg of dry KBr and the mixture is pressed in to a disk for spectral analysis.

3. RESULTS & DISCUSSION

3.1 Melting point Analysis

Various proportions of PVA-CHI-GEL films were prepared as shown in below table.2 and melting temperature is noted

.Table.2 Melting point of samples ABC

Sample	Gelatin (ml)	Melting Temperature (^oC)
I	13	199
II	11	201.5
III	9	204.5
IV	7	211
V	5	214.5
VI	3	215

From above table, it was found that melting point of the sample gradually increases. This indicates that with increase in gelatin concentration the melting point also increased.

3.2 Tensile Strength Test

The various proportions of PVA-CHI-GEL composite films were tested for tensile strength and readings were tabulated in below table.3.

Table.3 Tensile strength of PVA-CHI-GEL films

Sample	Maximum Load (N)	Tensile strength (MPa)	Elongation at break (%)
I	76.62	96.72	5.56
II	64.41	77.61	4.45
III	63.23	73.56	4.11
IV	49.21	58.37	3.72
V	39.34	52.31	3.22
VI	29.39	29.01	4.31

From above table it is clear that with increase in gelatin concentration the tensile strength and elongation increases.

3. 3 pH degradation
The films were submerged in pH 7.4 PBS buffer and degradation was noted in below table.4.

Table.4. pH degradation of PVA-CHI-GEL films

Sample	PVA-CHI blend (ml)	Gelatin (ml)	Film partial break (sec)	Total degradation (min)
I	2	13	150	8
II	4	11	180	8.5
III	6	9	230	10.5
IV	8	7	300	12
V	10	5	480	19.5
VI	12	3	580	31.5

From above table, it is clear that pH degradation of the films have higher resistance to degradation with higher concentration of PVA-CHI.

3.4 DC Resistivity of Optimized Ternary Blend
The samples (I-VI) were tested for dc resistance and their values were tabulated in below table.5.

Table.5. DC Resistance of PVA_CHI_GEL films

Sample	Resistance (MΩ)	Thickness (µm)	Resistivity (10^9Ω-m)
I	208	90	5.778
II	132	80	4.125
III	78	75	2.6
IV	45	77	1.461
V	42	65	1.615
VI	40	60	1.667

From above table, it is evident that the dc resistivity of films decreases with increase PVA-CHI. The resistivity decreases upto sample-4 and it saturates in samples 5 and 6.

3.5 AC resistivity of PVA-CHI-GEL films
The ac resistivity of samples is expressed interms of dielectric constant and is depicted in fig.2.

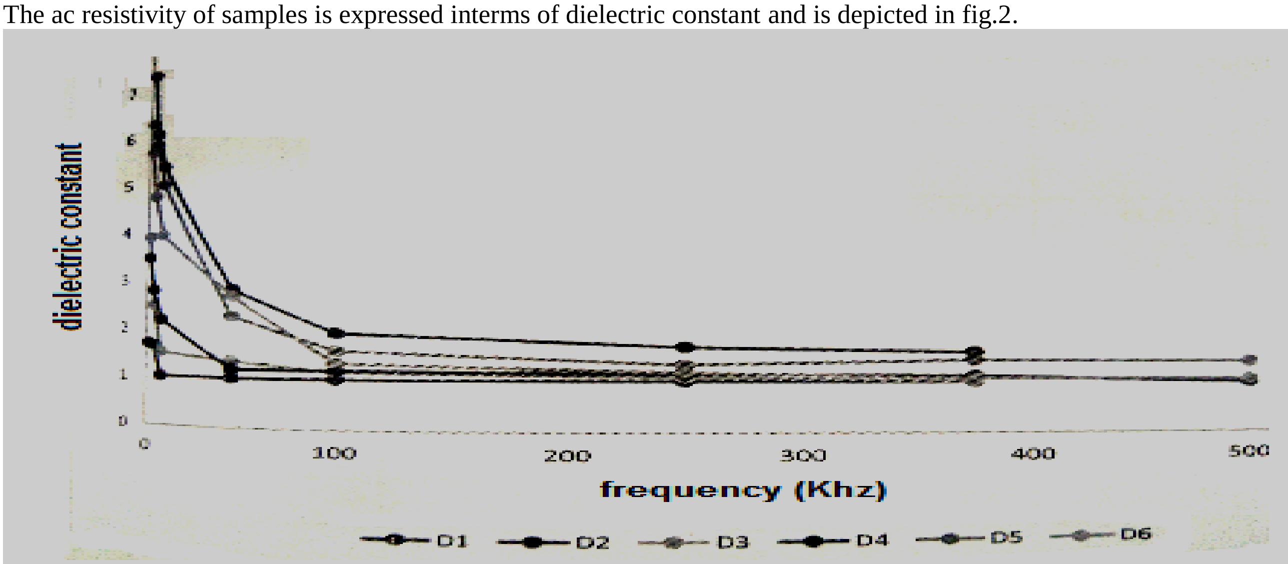

Fig.2 Dielectric constant of films with ac frequency

The dielectric constant of the films is inversely proportional to the ac frequency. The variation of the ac current flow through the samples verses ac frequency is shown in below fig.3.

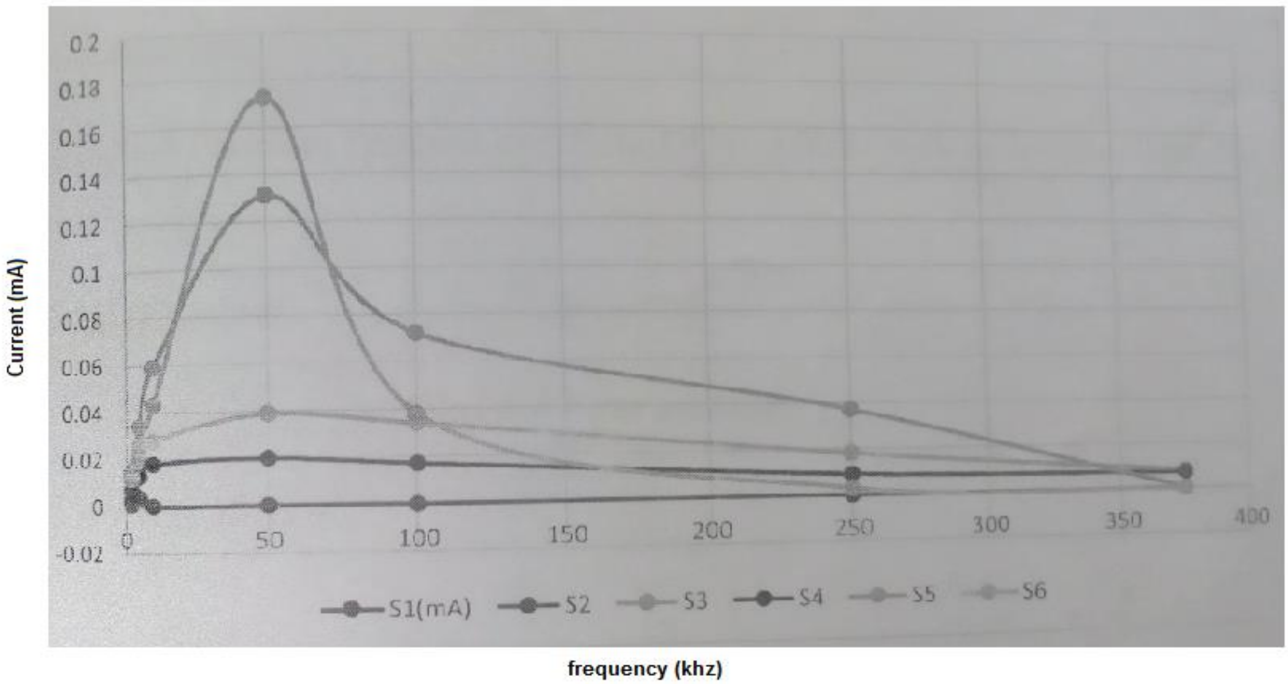

Fig.3 ac frequency X ac current

The figure shows an increase in ac current with frequency upto 50 khz and then decrease with frequency. This tells that the charge carriers will oppose current flows. Thus the developed films compositions have minimum ac resistance at 50 khz.

3.6 FTIR Analysis

The FTIR spectra are shown in below fig.4.

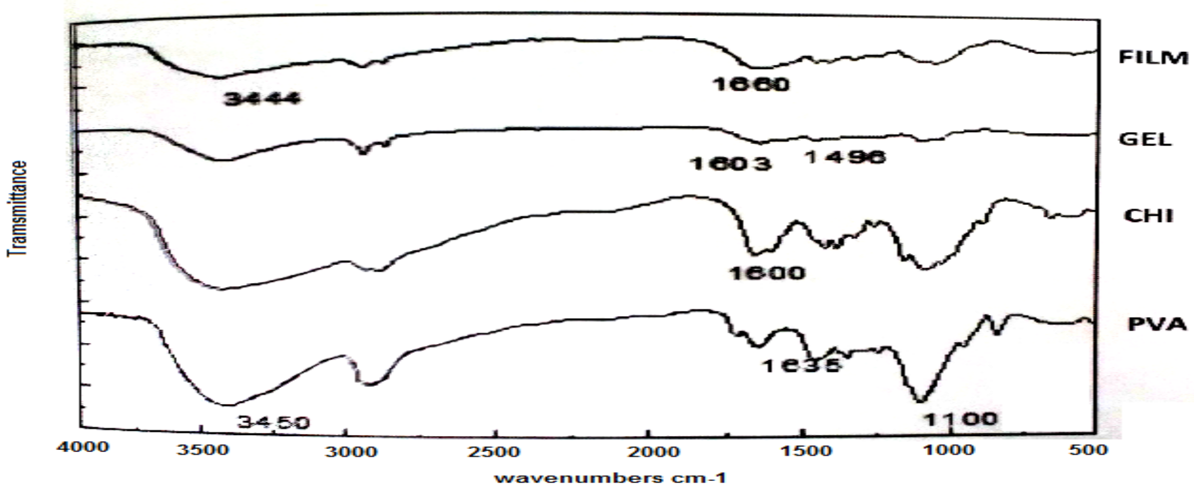

Fig.4 FTIR spectra of films

From the spectra, we observe that the band at 3450cm-1 corresponds to OH stretching vibration of PVA hydroxyl group. The band at 1635 cm-1 corresponds to CO stretching of the acetate group of

PVA. The band at 2910 cm-1 corresponds to stretching vibration of backbone aliphatic CH and 1100 cm-1 for CO stretching of PVA. In gelatin spectra, the band at 1603 cm-1belongs to CO and 1496 cm-1 is for NH bending. In Chitosan spectra the band at 1600 cm-1 represents CO. In PVA-CHI-GEL spectra we saw bands at 3444 cm-1 and at 1600 cm-1which confirms cross linking of the samples through hydrogen bonding interaction between the amino and hydroxyl groups.

3.7 DC conductivity of PVA-CHI-GEL-Carotenoid films

The sample-4 (optimized PVA-CHI-GEL) is doped with eight different amounts of carotenoid solution to study their characteristics as per below table.6.

Table.6 DC resistivity of PVA-CHI-GEL-Carotenoid films

Sample ID	Carotenoid (ml)	PVA-CHI-GEL (ml)
I	0	4
II	0.5	3.5
III	1.0	3.0
IV	1.5	2.5
V	2.0	2.0
VI	2.5	1.5
VII	3.0	1.0
VIII	3.5	0.5

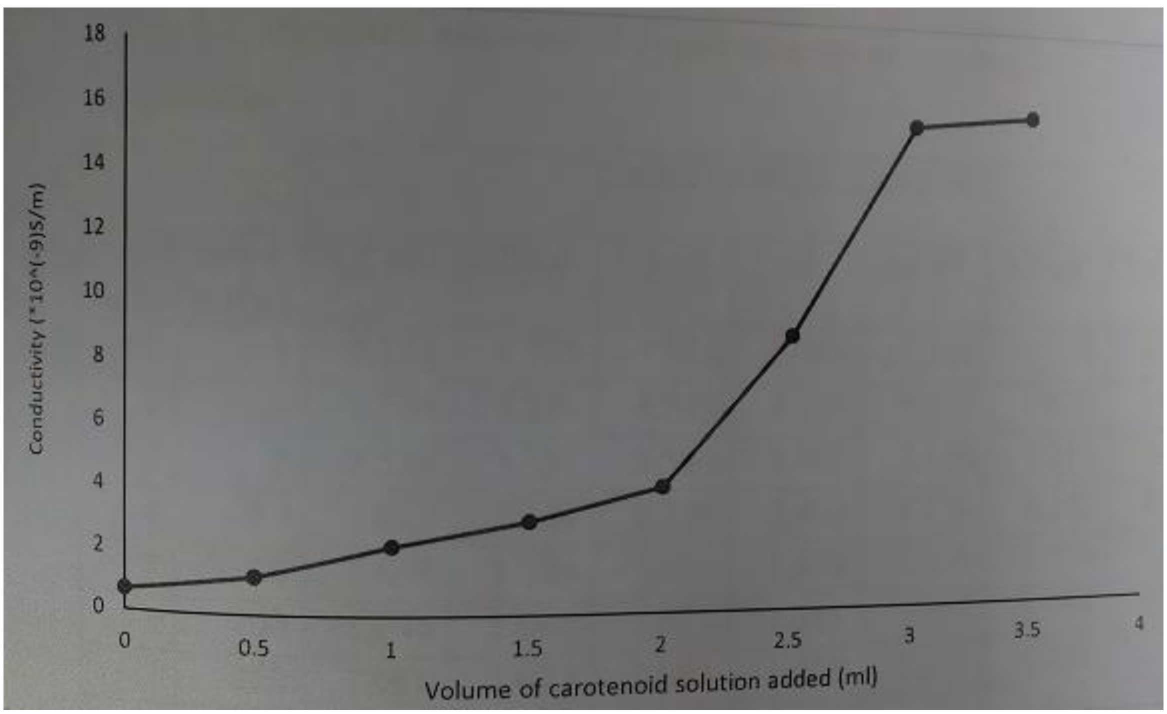

Fig.5. DC resistance of PVA-CHI-GEL-Carotenoid films

The DC conductivity of doped samples was found to be increased with carotenoid concentration. This is due to the contribution of π-electrons in carotenoid contributes to the electric current flow through the film.

3.8. AC conductivity of PVA-CHI-GEL-Carotenoid films

The ac conductivity of PVA-CHI-GEL-Carotenoid films is estimated interms of its dielectric constant against ac frequency as shown in table.7.

Table.7. Dielectric constant of PVA-CHI-GEL-Carotenoid films against ac frequency

		Carotenoid Amount (ml)						
S. No.	Frequency (khz)	0.5	1.0	1.5	2.0	2.5	3.0	3.5
I	1	6.183	6.775	7.055	7.786	11.111	11.38	13.11
II	2	3.564	5.274	4.851	7.275	9.830	10.40	11.25
III	5	2.553	3.551	3.390	2.901	7.850	7.216	7.216
IV	10	1.813	2.244	2.343	2.403	4.270	4.292	4.292
V	50	1.078	1.104	1.114	1.959	1.379	1.376	1.382
VI	100	1.046	1.065	1.082	1.361	1.189	1.194	1.194
VII	250	1.088	1.077	1.093	1.358	1.231	1.258	1.258
VIII	375	1.127	1.172	1.238	1.456	1.398	1.398	1.39

From the table.7 it can be seen that the dielectric constants of the samples increases with carotene concentration and the dielectric nature was found to saturate at between 3.0 to 3.5 ml of carotenoid. Thus it can be concluded that 3 ml of carotene extract is the optimal doping volume to get maximum ac electrical conductivity in the sample.

4. CONCLUSION

In my study, I conclude that the ternary blend film made in the ratio 7:8 of 5% gelatin and 2.5% PVA-Chitosan was found to be optimal interms of melting point, pH degradation, Tensile strength and electrical conductivity. This optimized composite possesses conductivity of $16x10^{-9}$ S/m which is in the range of semiconductors. Hence PVA-CHI-GEL-Carotenoid composite will have potential medical applications.

REFERENCES

[1] S. Lofty and Y.H.A Fawzy. "Characterization and enhancement of the electrical performance of radiation modified poly (vinyl) alcohol/gelatin copolymer films doped with carotene". Journal of Radiation Research and Applied Sciences, volume 7, pages 338-345, 2014.

[2] Hossam M Said. "Development of films based on poly (vinyl alcohol)/gelatin blends cross linked by Electron Beam Irradiation". Arab Journal of Nuclear Science and Applications, volume 46(5), pages 70-78, 2013.

[3] Gamal S. El Bahy, El-Sayed M. El-Sayed, Abdel Aziz Mahmoud and Noha M. Gweily. "Preparation and Characterization of poly vinyl alcohol / gelatin blends". Journal of Applied Sciences Research, volume 8(7), pages 3544-3551, 2012.

[4] Nazar Mohammad Ranjha, Samuillah Khan. "Chitosan/poly(vinyl alcohol) based hydrogels for biomedical applications: A Review". Journal of Pharmacy and Alternative medicine, volume 2, pages 30-41, 2013.

[5] Cheng-Ho Chen, Fang-Yu Wang, Ching-Feng Mao, Wei-Tung Liao and Ching-Dong Hsich. "Studies of Chitosan:II. Preparation and characterization of Chitosan/poly(vinyl alcohol)/gelatin ternary blend films". International Journal of biological macromolecules, volume 43, pages 37-42, 2008.

[6] Lihong Fan*, Huan Yang, Jing Yang, Min Peng and Jin Hu. "Preparation and characterization of Chitosan/gelatin/PVA hydrogel for wound dressing". Carbohydrate Polymers, volume 146, pages 427-434, 2016.

[7] G Leatherman, E N Durantini, Gust T A, Moore, A L Moore, S Stone, Z Zhou, P Rez, Y Z Liu and S M Lindsay. "Carotene as a Molecular Wire: Conducting Atomic Force Microscopy". Journal of Physical Chemistry B, volume 103, pages 4006-4010, 1999.