Bioactive constituents of *Centrosema pubescens* Benth. inhibitory to *Culex quinquefasciatus* larvae

Mary Angel D. Javier

Presenter

Department of Chemistry, College of Science, Tarlac State University

Gregorio Romulo Blvd., San Vicente, Tarlac City (Tarlac), Philippines

Project description

❑ Mosquito borne diseases are prevalent

- 100 countries across the world
- 700,000 people is infected every year

World Health Organization. 1996. Report of the WHO informal consultation on the evaluation on the testing of insecticides, CTD/WHO PES/IC/96.1. Geneva: WHO;. p. 69.

❑ Disease burden, death, poverty, and social debility in tropical countries

• **Malaria**	• **Filariasis**
• **Dengue**	• **Japanese encephalitis**
• **Yellow fever**	

Gubler, D.J. 1998. Emerg Infect Dis., 4, 442–450.

Project description

- 279 species, subspecies and varieties of mosquitoes recorded.

Cagampang-Ramos, A., McKenna, R. and Pinkovsky, D.D. 1985. A List of Philippine Mosquitoes (Diptera: Culicidae). Mosquito Systematics Vol. 17(l)

- Herbal medicines have been used for decades and centuries for curing diseases has been documented.

E. Quisumbing. 1978. Medicinal Plants of the Philippines. Katha, Caloocan City, pp. 517-8.

Project description

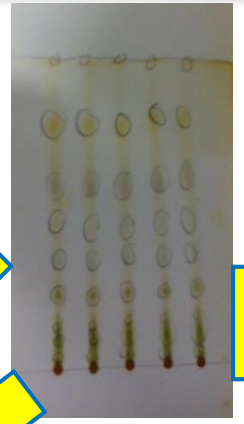
- Larvicidal potential of the crude ethanolic extracts of *Centrosema pubescens* Benth.

- ✓ 2nd Symposium on Health Research Projects in Region III, May 2015
- ✓ Global Health Forum, August 2015
- ✓ BMJ Open 5 Suppl. 1

Objectives

- Extract and isolate the bioactive components from the ethanolic extracts of the *Centrosema pubescens* Benth.against *Culex quinquefasciatus* larvae

Methodology



http://www.yanachemodities.com/

http://www.buchi.com/en/products/laboratory-

http://www.dlsu.edu.ph/conferences/dlsu_research_congress/2014/_pdf/proceedings/FNH-III-022-ft.pdf

http://npic.orst.edu/factsheets/malatech

$$\text{Percentage Larval Mortality} = \frac{\text{Total number of dead mosquito larvae}}{\text{Total number of mosquito larvae used}} \times 100$$

Results

Table 1. Percent mortality of *C. pubescens* B. crude ethanolic extract against Cx larvae

Test Concentration (ppm)	Percent Mortality 24 hours exposure	Percent Mortality 48 hours exposure
25	0	0
50	0	0
75	0	0
100	0	0
250	0	0
500	0	0
1000	0	0
2000	0	0
3000	0	0
4000	9	9
5000	18	17
6000	19	21
7000	30	32
8000	48	48
9000	66	68
10000	70	71
Distilled water	0	1
Malathion	100	100

Results. . . .

Table 2. Percent mortality of *C. pubescens* B. concentrate ethanolic extract against Cx larvae

Test Concentration (ppm)	Percent Mortality 24 hours exposure	Percent Mortality 48 hours exposure
25	0	0
50	0	0
75	0	0
100	0	0
250	0	1
500	0	1
1000	3	7
2000	11	18
3000	14	23
4000	21	30
5000	31	43
6000	78	90
7000	80	90
8000	79	92
9000	83	94
10000	88	100
Distilled water	0	1
Malathion	100	100

Results. . . .

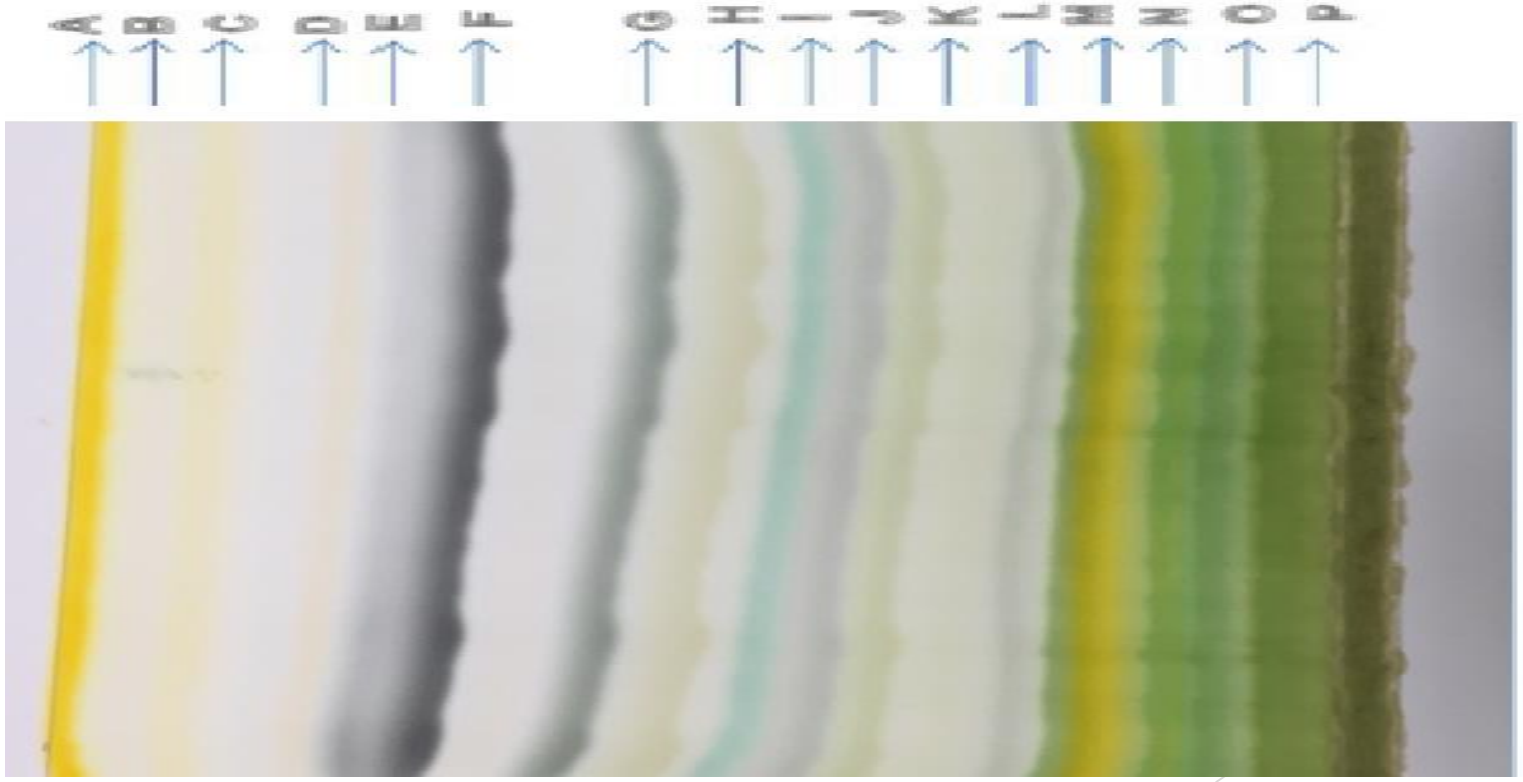
Table 3. LC_{50}/LD_{50} and LD_{90}/LC_{90} using *Cx* larvae as test organism.

Treatment	LD_{50}/LC_{50}		LD_{90}/LC_{90}	
	24 hours	48 hours	24 hours	48 hours
Crude extract	8090.06 ppm	8053.78 ppm	14879.68 ppm	14370.34 ppm
Concentrate extract	4808.39 ppm	3396.25 ppm	12022.64 ppm	10299.16 ppm

Results. . . .

Table 4. TLC profile of the crude ethanolic extract concentrate of *C. pubescens* B.

Isolated Compound	A	B	C	D	E	F	G	H	I	J	K	L	M	O	P	Q	R	S
Rf values	0.95	0.91	0.82	0.81	0.67	0.62	0.54	0.46	0.43	0.34	0.32	0.26	0.26	0.22	0.32	0.16	0.09	0.04



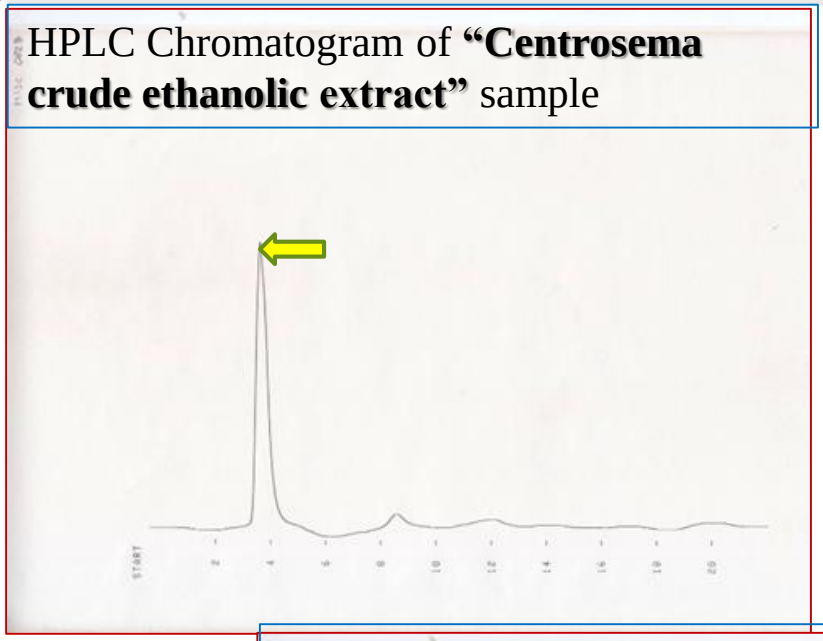
Results. . . .

Tables 5. Larval mortality of the isolated compounds in minutes against Cx larvae.

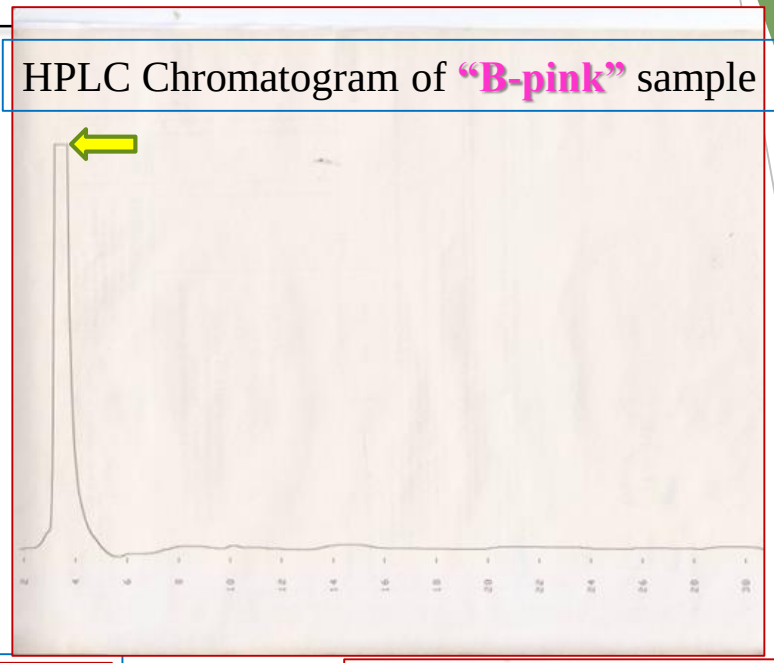
Isolated Compounds	Time of Death (minute)	
	1000 ppm	10000 ppm
I	149	13
A	208	18
C	338	29
B	771	87
J	869	112
F	1109	121
E	1150	156
K	1176	208
D	1357	218
G	1361	294
L	1405	333
H	1413	345
M	1430	1192

Results. . . .

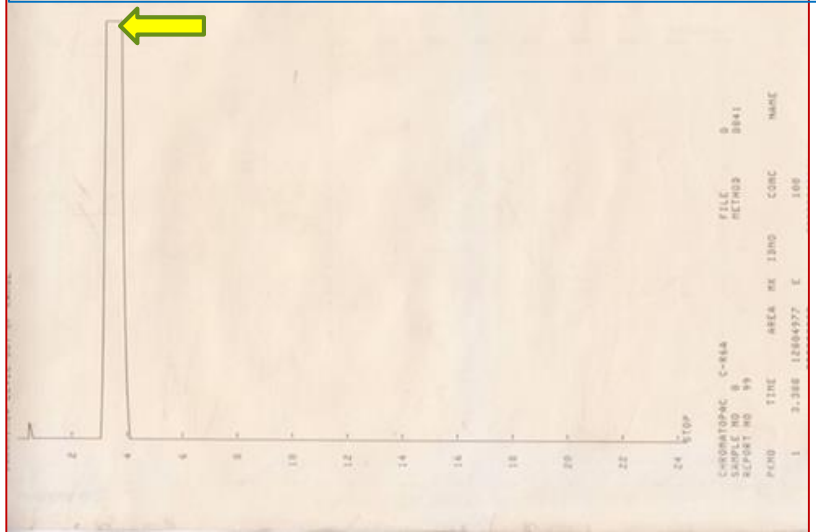
HPLC Chromatogram of **"Centrosema crude ethanolic extract"** sample



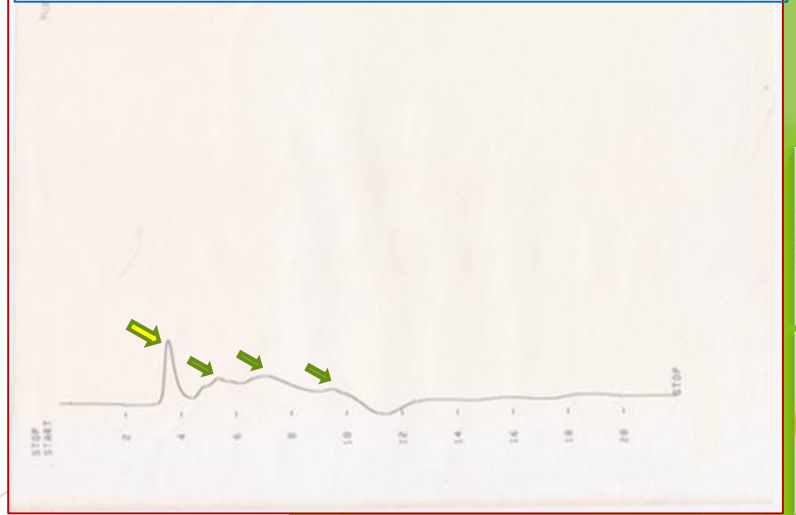
HPLC Chromatogram of **"B-pink"** sample



HPLC Chromatogram of **"A-Yellow"** sample

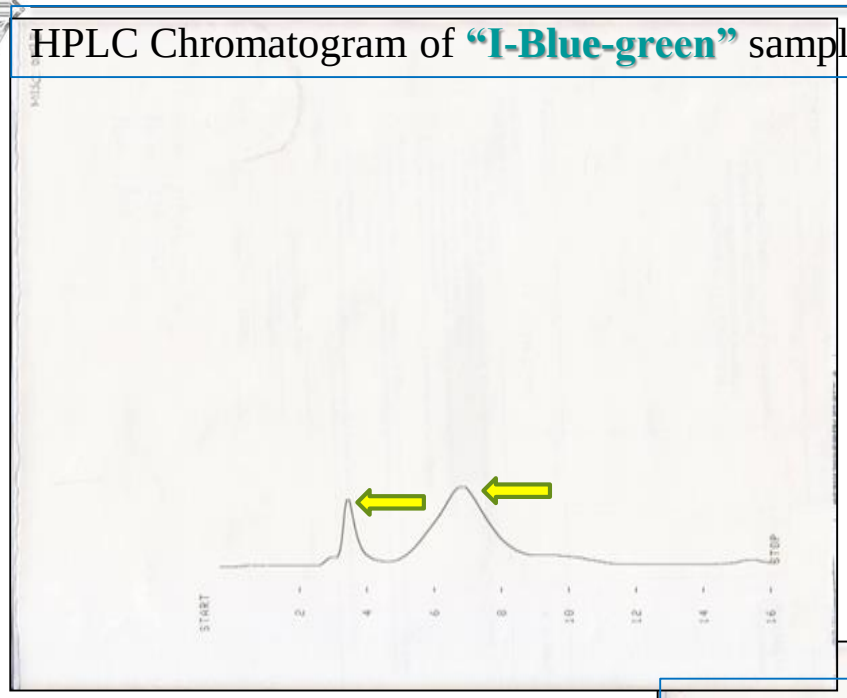


HPLC Chromatogram of **"C-Yellow"** sample

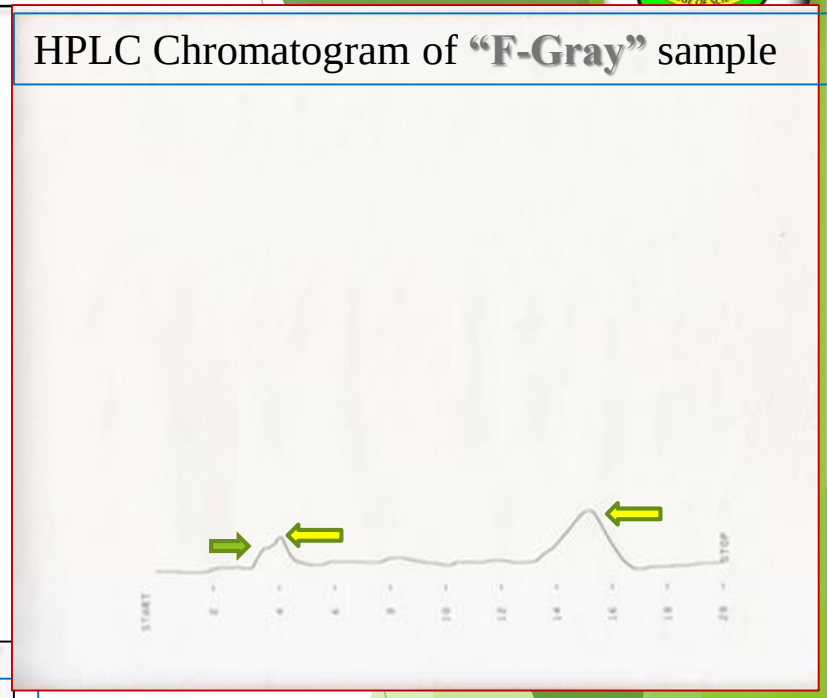


Results. . . .

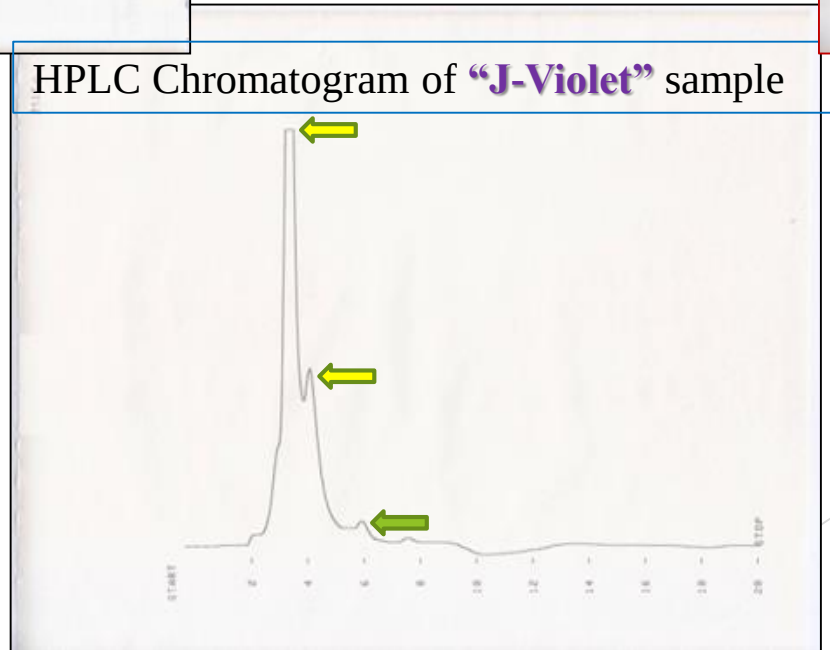
HPLC Chromatogram of **"I-Blue-green"** sample



HPLC Chromatogram of **"F-Gray"** sample

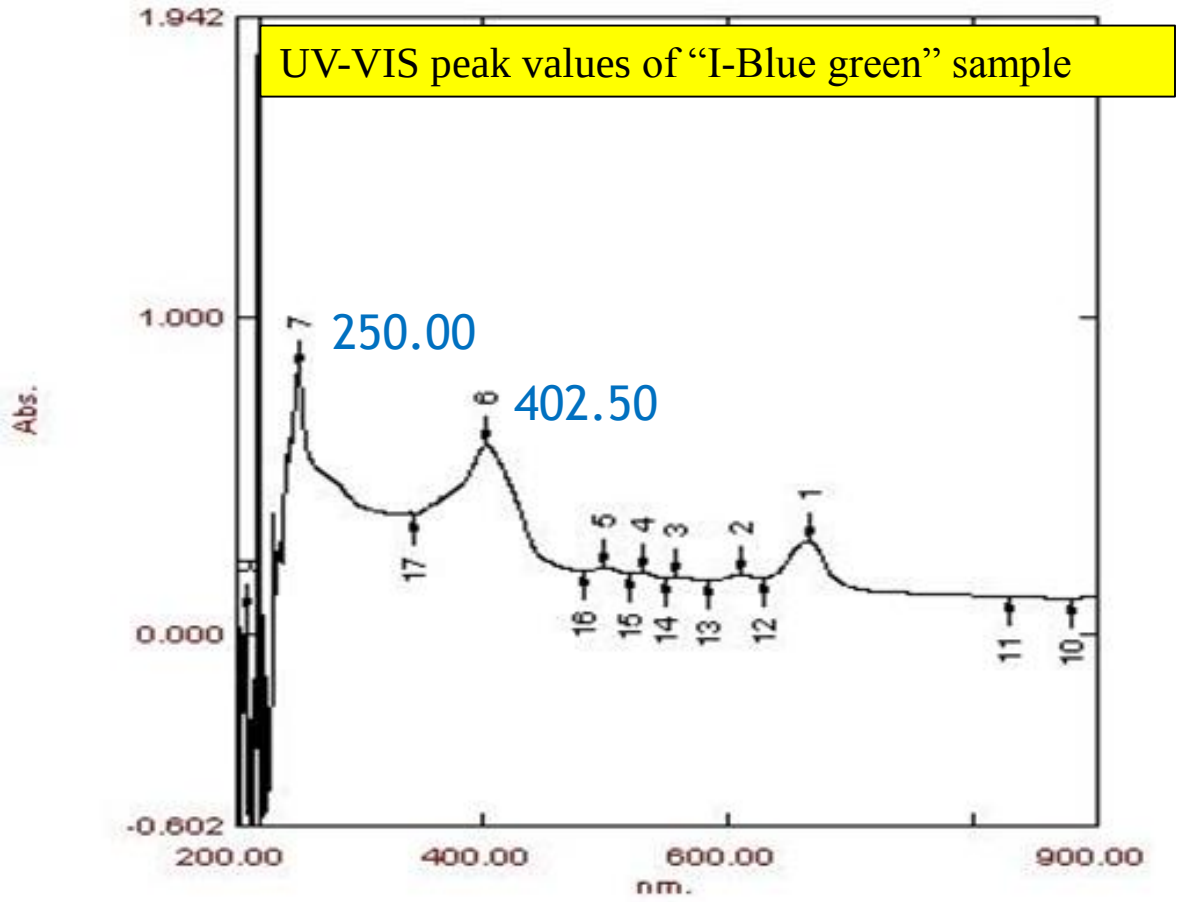


HPLC Chromatogram of **"J-Violet"** sample

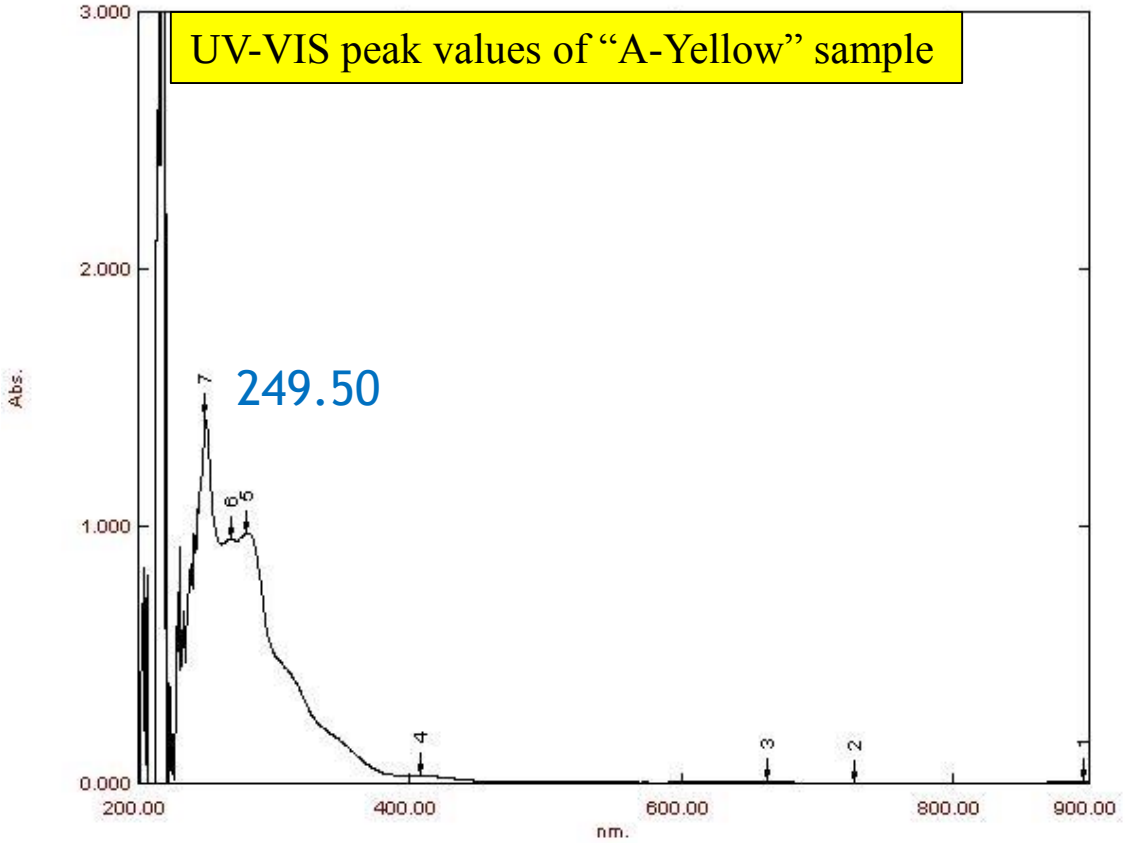


Results. . . .

UV-VIS peak values of "I-Blue green" sample

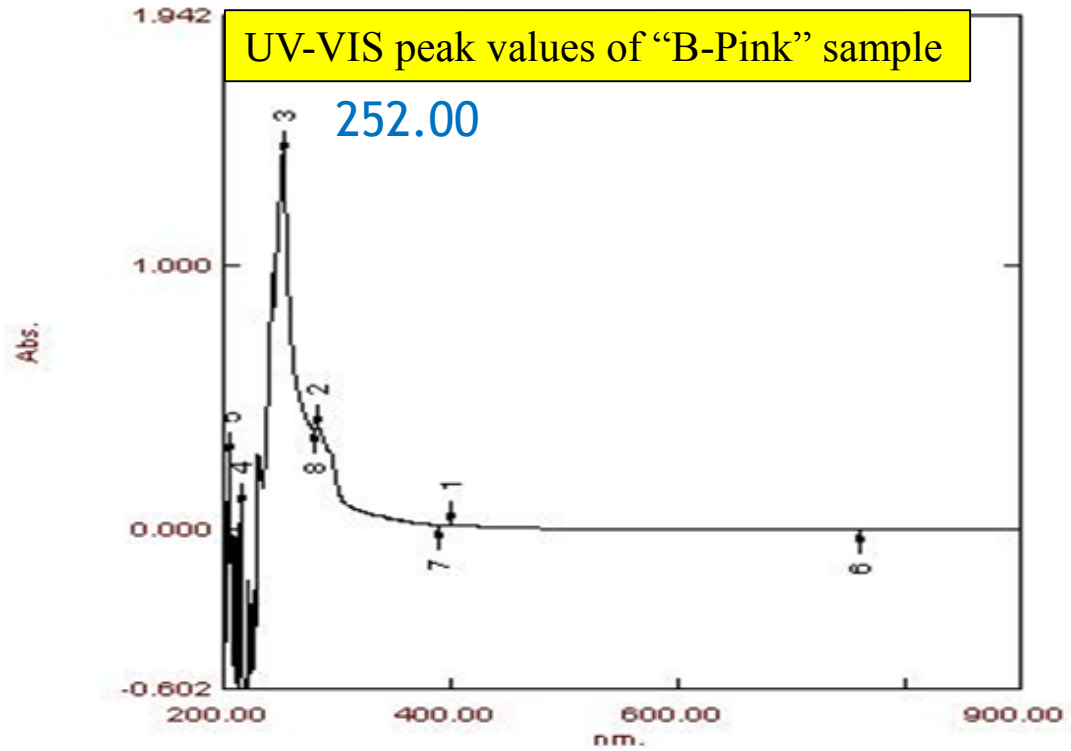


UV-VIS peak values of "A-Yellow" sample

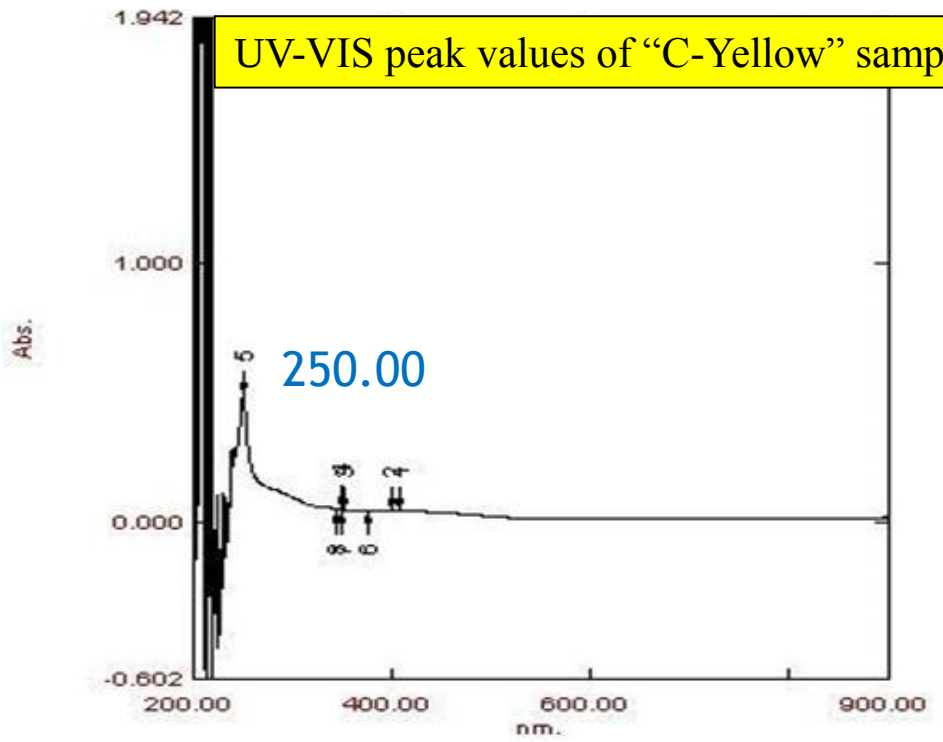


Results. . . .

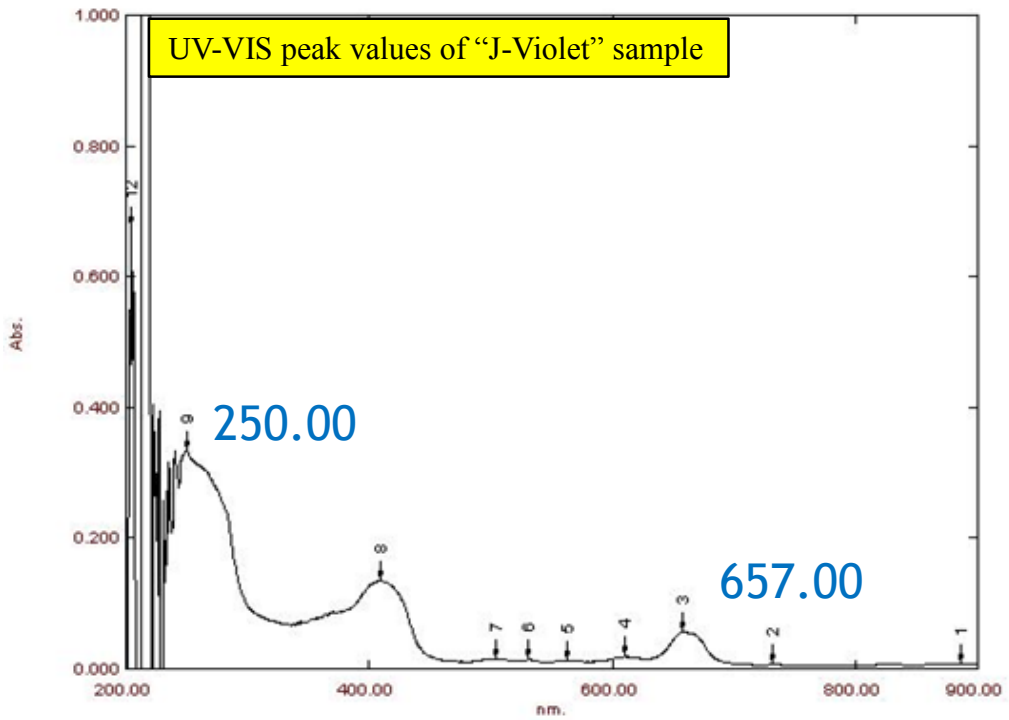
UV-VIS peak values of "B-Pink" sample

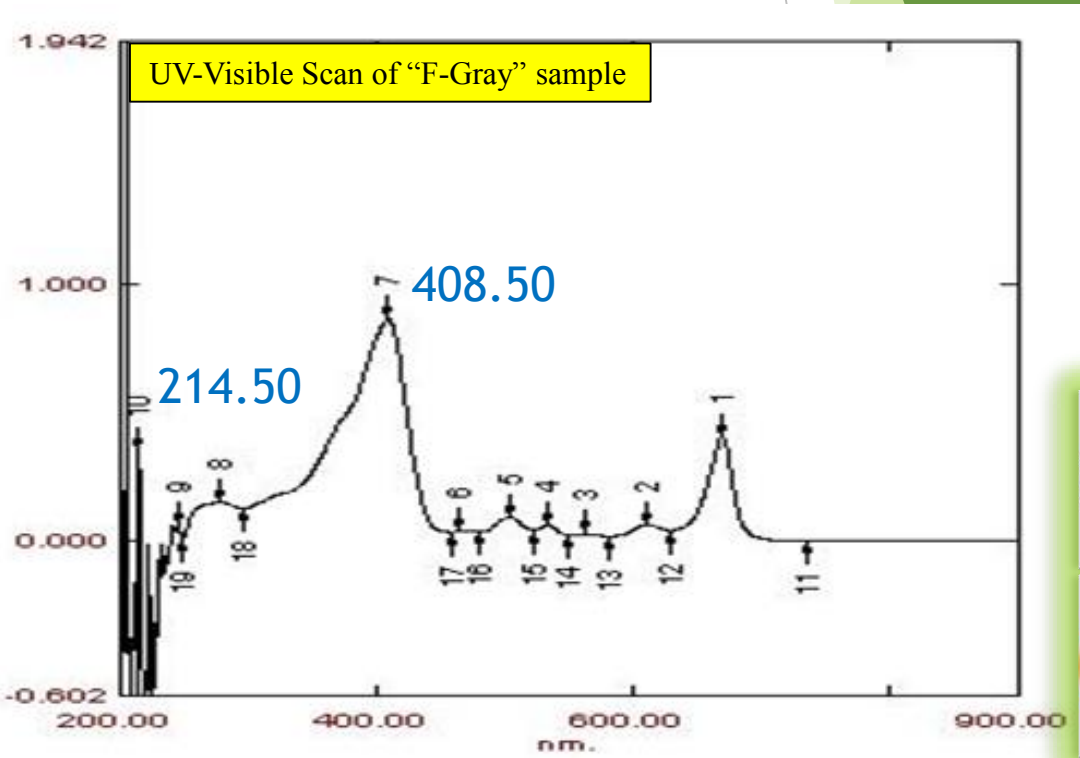


UV-VIS peak values of "C-Yellow" sample



Results. . . .

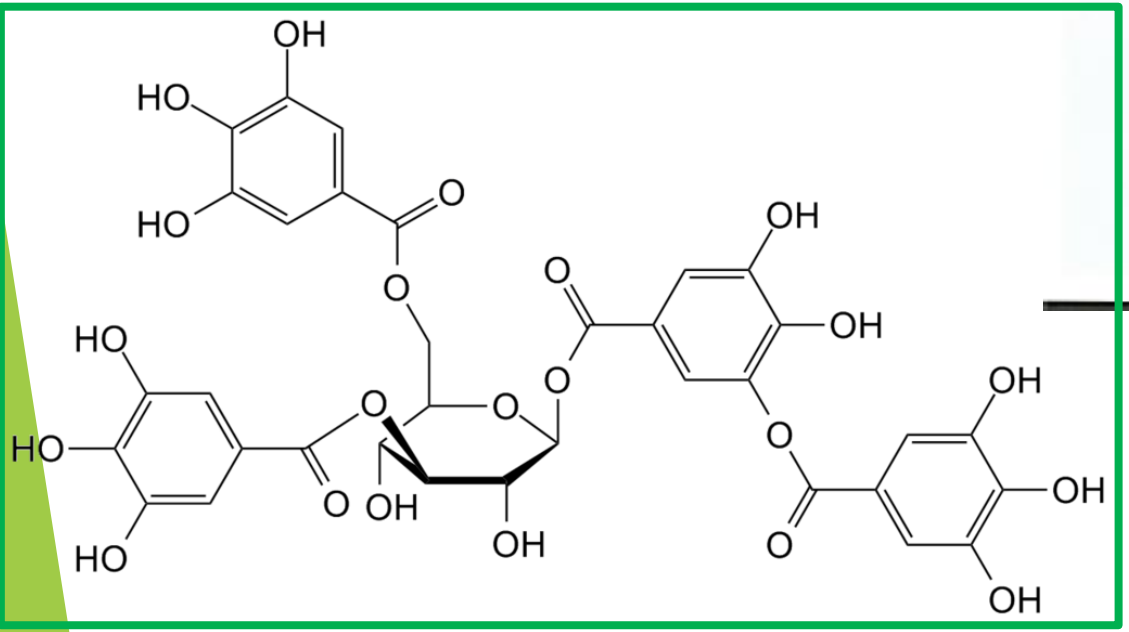

UV-VIS peak values of "J-Violet" sample
1.000
0.800
0.600
0.400
0.200
0.000
Abs.
250.00
657.00
200.00
400.00
600.00
800.00
900.00
nm.

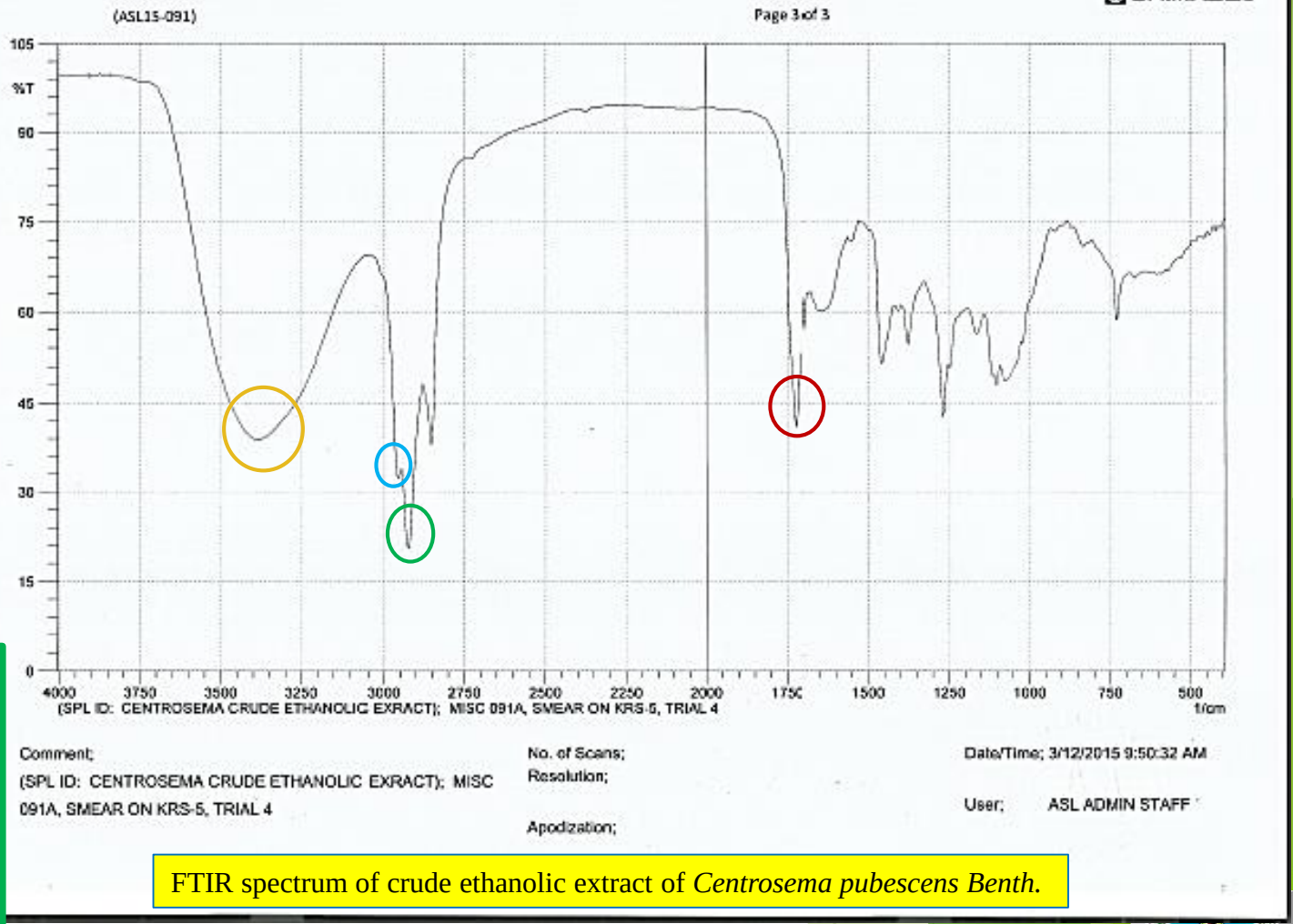

UV-Visible Scan of "F-Gray" sample
1.942
1.000
0.000
-0.602
Abs.
408.50
214.50
200.00
400.00
600.00
900.00
nm.

Results. . . .

Table 5.18. FTIR peak values of crude ethanolic extracts concentrate of *Centrosema pubescens* Benth.

Frequency (cm^{-1})	Functional groups
1725	C=O
2910	sp^3 C-H stretch
2950	sp^2 C-H stretch
3375	O-H stretch





FTIR spectrum of crude ethanolic extract of *Centrosema pubescens Benth.*

Solanine is a glycoalkaloid poison found in species of the Solanaceae family.

Conclusion

- LC_{50}/LD_{50} and LC_{90}/LD_{90} values of the ethanolic extracts reveal inhibitory activity against Culex larvae.
- Compounds I, A, C, B, and J were isolated through TLC; Compound I shows the most appreciable larvicidal activity.
- Spectroscopic analyses confirmed the presence of alcohols and aromatic compounds which is responsible for its inhibitory against Culex larvae.

Recommendation

- **Structure determination of the isolated components from crude ethanolic extract concentrate using Nuclear Magnetic Resonance (NMR) spectroscopy, Mass spectrometry and Melting Point Determination.**

Bioactive constituents of *Centrosema pubescens* Benth. inhibitory to *Culex quinquefasciatus* larvae