

A Quantitative Analysis of Chlorophyll Depletion by Aspirin and Paracetamol in *Oryza sativa*

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Physiologically active organic molecules (PAOM) like drugs can interact and involve in plant's physiology and Biochemical system. The extent of absorption and interaction of these PAOM is a function of their structure and polarity of functional groups. The present investigation deals with impact of simple phenolic drugs like Aspirin and Paracetamol (Acetaminophen) on depletion of chlorophyll content in *Oryza sativa*.

MATERIALS AND METHODS

Culturing the Rice plants with Drug Treatment

The pot experiment was conducted in silty loam soil with the alkali-hydrolysable N, P₂O₅, K₂O and 200, 400, 1000 and 2000ppm of Aspirin and Paracetamol separately. The contents are thoroughly mixed. Ten kilogram of soil was placed in each pot (26cm in diameter and 30 cm in height). The pre grown rice plants (11th leaf plant) introduced into above thoroughly mixed soil submerged in water. Fertilizers were applied as a basal dressing after 3 days (2.0g N, 1.0g P₂O₅, and 1.0g K₂O). These pots were flooded with a water layer of 3cm during whole growth period.

The samples are harvested for 10 days in photic chamber and measured for fluctuations in chlorophyll contents.

Extraction and Estimation of chlorophyll pigments:

Arnon (1949) method was adopted for estimation of chlorophyll pigments. Leaves of *Oryza sativa* were washed with distilled water and dried. Take 200mg of leaf material

and macerated in 2ml of 80% acetone and separate the extract using mechanical centrifuge repeat the procedure for 3-4 times with same material until till white pellet was obtained. Make up the extract for 25ml with 80% acetone. Quantitative analysis was done by spectrophotometrical method. Optical density (O.D.) values are measured by using Systronics 106 Automat instrument at λ_{max} of 663nm, 645nm & 480nm for calculating the chl-a and chl-b pigments.

Calculations:

Chlorophyll 'a' ($\mu\text{g}\cdot\text{g}^{-1}$ f.w.)

$$((\text{O.D.}663*12.7)-(\text{O.D.}645*2.69))*V/1000xW$$

Chlorophyll 'b' ($\mu\text{g}\cdot\text{g}^{-1}$ f.w.)

$$((\text{O.D.}645*22.9)-(\text{O.D.}663*4.68))*V/1000xW$$

Total Chlorophyll ($\mu\text{g}\cdot\text{g}^{-1}$ f.w.)

$$((\text{O.D.}645*20.2)-(\text{O.D.}663*8.02))*V/1000xW$$

f.w. : Fresh Weight.

Estimation:

Sample Collected From Control Paddy Plant

	Chlorophyll-a ($\mu\text{g}\cdot\text{g}^{-1}$ f.w)	Chlorophyll-b ($\mu\text{g}\cdot\text{g}^{-1}$ f.w)	Total chlorophyll ($\mu\text{g}\cdot\text{g}^{-1}$ f.w)
Control Plant			
A	49.803	49.282	98.9283
B	49.513	49.1235	98.606
C	49.58	48.551	98.101

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Average	49.632	48.9855	98.5451
standard deviation	0.151832144	0.38454291	0.416998717
standard error of the mean	0.087	0.222	0.24
	49.632±0.08	48.98±0.22	±0.24 98.54
Sample Collected From Paddy Plant cultured in 200ppm Acetaminophen			
A	23.333	24.2646	47.583
B	23.072	25.456	48.512
C	23.41	24.126	47.522
Average	23.27166667	24.61553333	47.87233333
standard deviation	0.177150595	0.731157065	0.554806573
standard error of the mean	0.102	0.422	0.32
	0.102 ± 23.27	±0.42 24.61	±0.32 47.87
Sample Collected From Paddy Plant cultured in 400ppm Acetaminophen			
A	22.425	23.723	46.133
B	22.261	23.564	45.811
C	22.134	23.611	45.731
Average	22.27333333	23.63266667	45.89166667
standard deviation	0.145891512	0.081684352	0.21279411
standard error of the mean	0.084	0.047	0.122
	±0.084 22.27	±0.04 23.63	±0.12 46.13
Sample Collected From Paddy Plant cultured in 1000ppm Acetaminophen			
A	6.575	9.188	15.758
B	6.221	9.1	15.316
C	6.552	8.868	15.415
Average	6.449333333	9.052	15.49633333
standard deviation	0.198076585	0.165311827	0.2319533
standard error of the mean	0.11	0.095	0.133922229
	±0.11 6.44	±0.09 9.05	±0.13 15.49
Sample Collected From Paddy Plant cultured in 2000ppm Acetaminophen			
A	3.249	6.47	9.716
B	3.212	6.076	9.474
C	3.178	7.592	10.766
Average	3.213	6.712666667	9.985333333
standard deviation	0.035510562	0.786593499	0.686819724
standard error of the mean	0.02	0.454	0.396547185
	±0.02 3.213	±0.45 6.71	±0.39 9.98
(The values are mean±S.E(n=3)			

Sample Collected From Paddy Plant cultured in 200ppm Aspirin			
Subtypes of Chlorophyll pigments	(.Chlorophyll-a (µg.g ⁻¹ f.w	Chlorophyll-b (µg.g ⁻¹ f.w)	Total chlorophyll (µg.g ⁻¹ f.w)
A	97.13985	83.2775	180.363
B	95.89675	83.5165	179.359
C	97.82865	82.5855	180.36
Average	96.95508333	83.1265	180.0273333
standard deviation	0.979113601	0.483519389	0.578795589

standard error of the mean	0.565	0.279	0.334
	96.95±0.565	±0.279 83.12	±0.334 180
Sample Collected From Paddy Plant cultured in 400ppm Aspirin			
A	40.62375	33.7769	74.3784
B	40.3832	33.298	73.713
C	40.76045	34.1647	74.9027
Average	40.58913333	33.74653333	74.3313666
standard deviation	0.190992476	0.434147237	0.596242923
standard error of the mean	0.11	0.25	0.343
	40.58±0.11	33.74±0.25	±0.343 74.33
Sample Collected From Paddy Plant cultured in 1000ppm Aspirin			
A	30.66705	33.7215	64.368
B	30.0455	33.841	63.866
C	30.29575	34.30575	64.5715
Average	30.3361	33.95608333	64.2685
standard deviation	0.312733421	0.308658623	0.363122225
standard error of the mean	0.18	0.178	0.209
	30.33±0.18	33.95±0.176	64.26±0.206
Sample Collected From Paddy Plant cultured in 2000ppm Aspirin			
A	4.23265	5.3415	9.571
B	3.6111	5.461	9.069
C	4.273	4.998	10.486
Average	4.038916667	5.266833333	9.708666667
standard deviation	0.371048994	0.24036136	0.718461087
standard error of the mean	0.214	0.138	0.414
	4.038±0.214	5.26±0.138	9.70±0.414
(The values are mean±S.E(n=3)			

RESULTS AND DISCUSSION

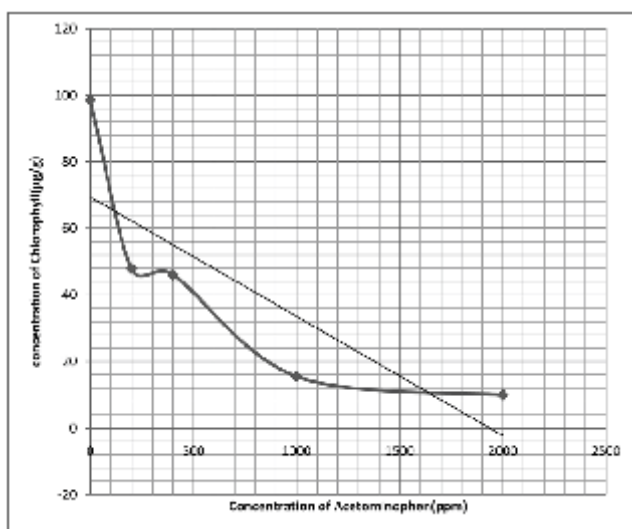


Fig1. Impact of Acetaminophen on total chlorophyll content.

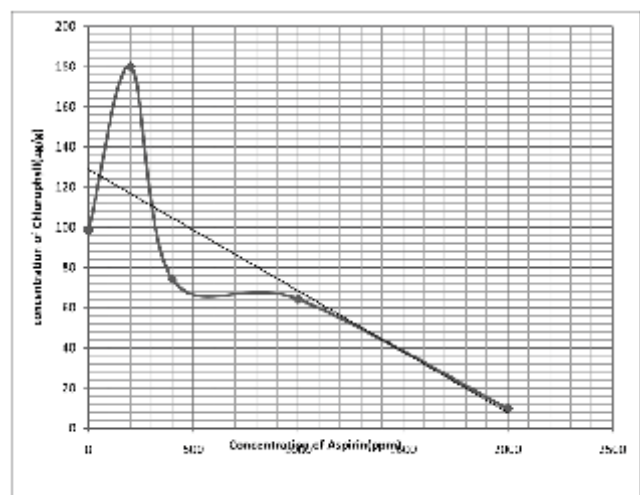


Fig2. Impact of Aspirin on total chlorophyll content.

There was mere research concerning the effects of pharmaceutical drugs on plant physiology. The current experiment clearly indicated that Aspirin and paracetamol shows incredible changes in physiology of *Oryza Sativa* in many aspects. This is due to either the interaction of these Physiologically Active Organic Molecules directly with phyto-hormones /Growth Regulators or itself acts as growth regulators. The above said drugs gradually degraded by soil bacteria and environmental factors into their phenolic monographs (4-amino phenol in case of paracetamol and Salicylic acid in case of Aspirin) which are more physiologically active as the polarity of functional group increases. This process slowly takes place hence the sequential assimilation by plants will not cause any chemical stress to the plants. In case of paracetamol (acetaminophen) gradual depletion of chlorophyll of all sub categories (chl-a,b) occurs with increase in concentration gradient. Where as in case of Aspirin these steep rise in chlorophyll content with 200ppm as its monograph i.e. Salicylic acid acts as physiological booster by exhibiting phyto-hormone activity but further increase in the dose will push the plant's physiology into chemical stress which leads to depletion in chlorophyll.

This experiment reveals that the Physiologically active organic molecules (PAOM) like drugs shows significant impact not only on animals but also on plants to various extents.

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