



**PONDICHERRY CENTRE FOR BIOLOGICAL SCIENCE AND
EDUCATIONAL TRUST**

Reg.No.2840/B4/2016 Pondicherry

GST IN: 33AADTP1541L1ZL

SUSTAINABLE RESEARCH FOR BETTERMENT OF MANKIND

CERTIFICATE OF ANALYSIS

PCBSET/2021-2022/RN00132_2

Date: 23/03/2022

User information: Mrs. J. Manonmani,
Research Scholar,
Annamalai University,
Annamalai Nagar, Chidambaram – 608002,
Tamil Nadu,
India

Sample information: Powder

Sample Code: C.A, G.A, S.A

Analysis: DPPH Assay

Date of sample received: 14/03/2022

Date of Analysis: 11/03/2022

Date of report: 12/03/2022

NITI Aayog



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MINISTRY OF MICRO, SMALL & MEDIUM ENTERPRISES
UDYAM REGISTRATION NUMBER - UDYAM-TN-31-0009450



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Methodology:

DPPH radical scavenging activity of extract was determined according to the method reported by Blois (1958). An aliquot of 0.5 ml of sample solution in methanol was mixed with 2.5 ml of 0.5 mM methanolic solution of DPPH. The mixture was shaken vigorously and incubated for 30 min in the dark at room temperature. The absorbance was measured at 517 nm using UV spectrophotometer. Ascorbic acid was used as a positive control. DPPH free radical scavenging ability (%) was calculated by using the formula.

$$\% \text{ of inhibition} = \frac{\text{absorbance of control} - \text{absorbance of sample}}{\text{absorbance of control}} \times 100.$$

Reference:

Blois MS. Antioxidant determinations by the use of a stable free radical. Nature. 1958;181:1199–1200. doi: 10.1038/1811199a0

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Results:

Sample code: C.A

Tested concentration($\mu\text{g/ml}$)	OD at 517nm(triplicate values)		
	500	0.3127	0.3121
250	0.3143	0.3158	0.3161
100	0.3170	0.3175	0.3185
50	0.3229	0.3223	0.3212
25	0.3242	0.3261	0.3249
Control	0.33	0.33	0.34

Tested concentration($\mu\text{g/ml}$)	% of Inhibition			Mean	Standard Deviation	Standard Error
	500	5.24	5.42			
250	4.76	4.30	4.21	4.42	0.29	0.17
100	3.94	3.78	3.48	3.74	0.23	0.13
50	2.15	2.33	2.67	2.38	0.26	0.15
25	1.76	1.18	1.55	1.49	0.29	0.17
Control	0.00	0.00	0.00	0.00	0.00	0.00

NITI Aayog



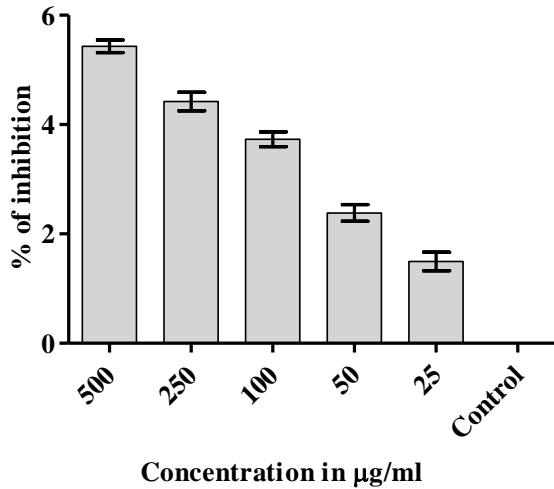
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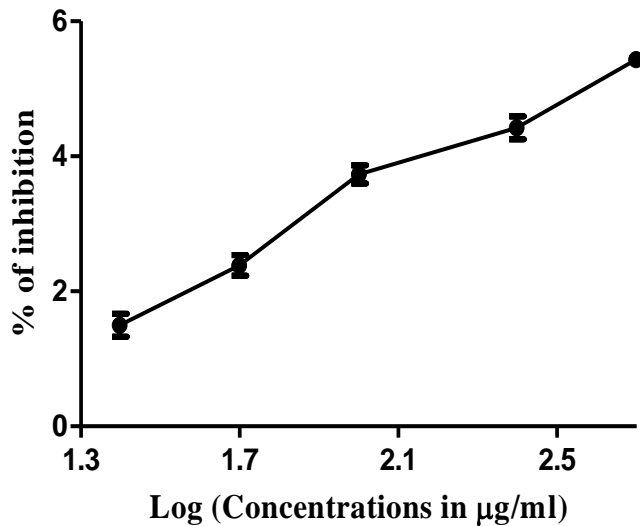
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IC50 Graph



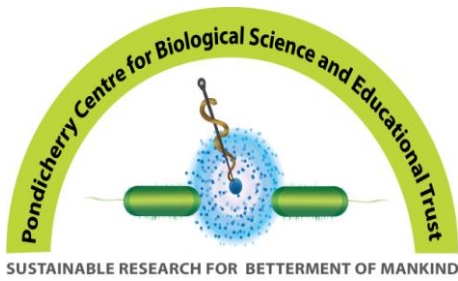
IC50 = >500 µg/ml

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Results:

Sample code: G.A

Tested concentration($\mu\text{g/ml}$)	OD at 517nm(triplicate values)		
	500	0.3155	0.3171
250	0.3191	0.3178	0.3183
100	0.3217	0.3224	0.3202
50	0.3242	0.3236	0.3231
25	0.3257	0.3274	0.3267
Control	0.33	0.33	0.34

Tested concentration($\mu\text{g/ml}$)	% of Inhibition			Mean	Standard Deviation	Standard Error
	500	4.39	3.91			
250	3.30	3.70	3.55	3.52	0.20	0.11
100	2.52	2.30	2.97	2.60	0.34	0.20
50	1.76	1.94	2.09	1.93	0.17	0.10
25	1.30	0.79	1.00	1.03	0.26	0.15
Control	0.00	0.00	0.00	0.00	0.00	0.00

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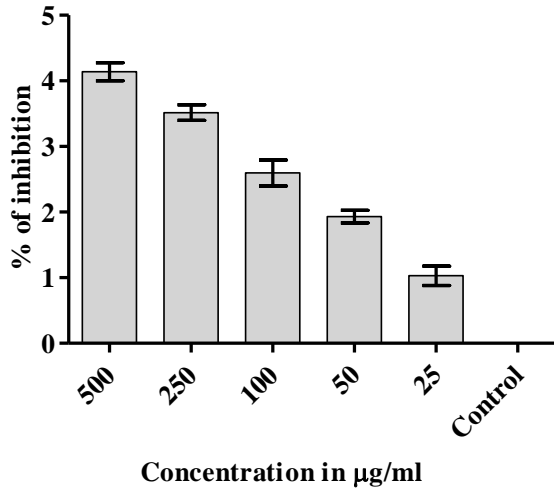
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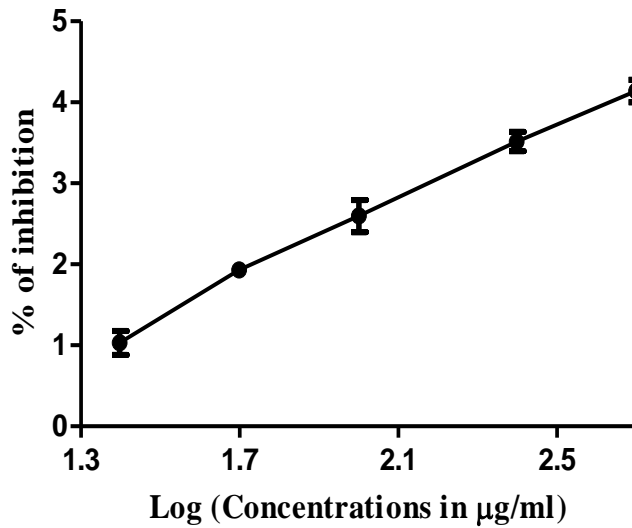
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IC50 Graph



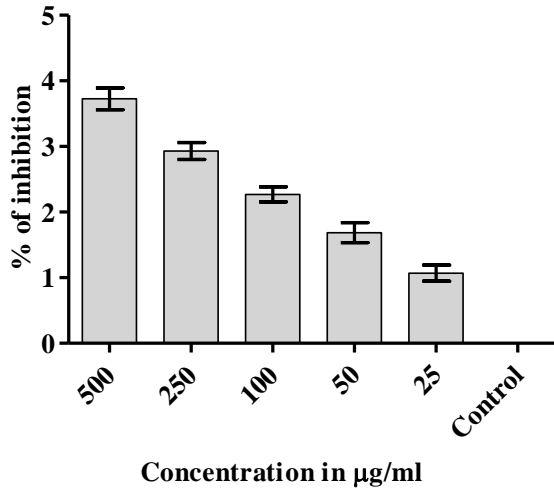
IC50 = >500 $\mu\text{g/ml}$

Results:

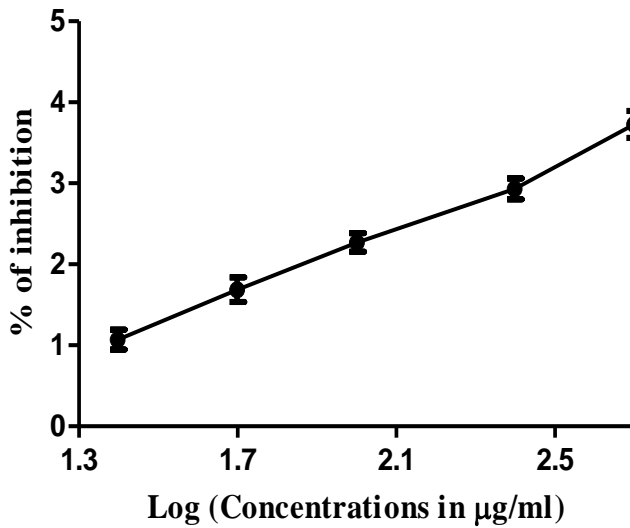
Sample code: S.A

Tested concentration($\mu\text{g/ml}$)	OD at 517nm(triplicate values)		
	500	0.3187	0.3176
250	0.3211	0.3203	0.3196
100	0.3219	0.3232	0.3224
50	0.3252	0.3246	0.3235
25	0.3271	0.3266	0.3257
Control	0.33	0.33	0.34

Tested concentration($\mu\text{g/ml}$)	% of Inhibition			Mean	Standard Deviation	Standard Error
	500	3.42	3.76			
250	2.70	2.94	3.15	2.93	0.23	0.13
100	2.45	2.06	2.30	2.27	0.20	0.11
50	1.45	1.64	1.97	1.69	0.26	0.15
25	0.88	1.03	1.30	1.07	0.21	0.12
Control	0.00	0.00	0.00	0.00	0.00	0.00



IC50 Graph



IC50 = >500 µg/ml

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Positive Control: Ascorbic acid

Tested concentration($\mu\text{g/ml}$)	OD at 517nm(triplicate values)		
5	0.169	0.171	0.174
10	0.037	0.033	0.032
20	0.032	0.029	0.028
40	0.026	0.028	0.025
50	0.024	0.022	0.024
Control	0.33	0.33	0.34

Tested concentration($\mu\text{g/ml}$)	% of Inhibition			Mean	Standard Deviation	Standard Error
5	48.82	48.24	47.36	48.14	0.73	0.42
10	88.91	90.03	90.39	89.78	0.77	0.45
20	90.45	91.21	91.52	91.06	0.55	0.32
40	92.12	91.61	92.33	92.02	0.37	0.22
50	92.73	93.21	92.76	92.90	0.27	0.16
Control	0.00	0.00	0.00	0.00	1.48	0.86

NITI Aayog



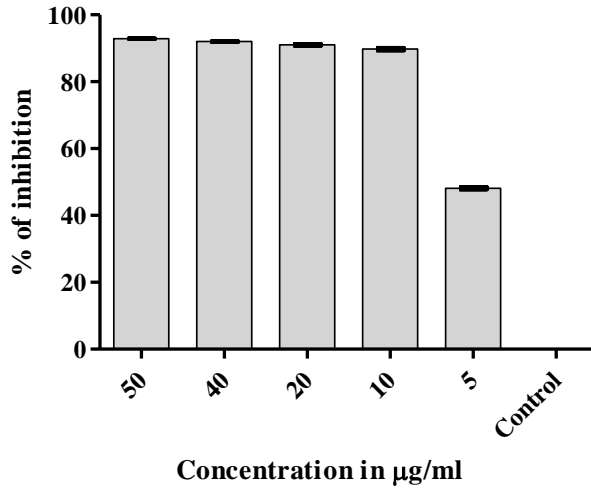
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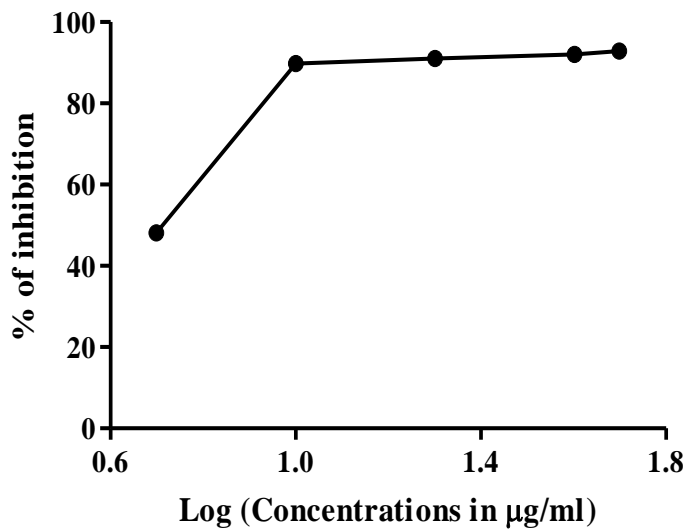
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IC50 Graph



IC50 = 5.064 ± 0.9 µg/ml