Results

The experiments were conducted to study the germination parameters, growth, stomatal structures, nodulation, root knot nematode growth, productivity and some biochemical changes in cowpea (*Vigna unguiculata* (L.) Walp.), under the effect of different concentrations of extracts of four parts of Parthenium plant, at three different stages of growth. The results obtained from the observation taken during the study are produced in Tables A to E and 1 to 20; Figures A, B and 1 to11; Plates No.1 to 48.

Section A

Seed Germination and Seedling Growth

The screening experiment was conducted to observe the effect of different concentrations of Parthenium extracts on seed germination parameters and seedling growth of cowpea variety Gomati, under lab condition in Petridishes. The results are presented in Tables A and B, Figures A and B and Plate No. 1 and 2.

Days of Maximum Germination and G%

The observation revealed that all the concentrations initiate early germination, except 200% of RE as compared to control (Table-A; Plate No.-1). In untreated control set, the maximum germination was recorded (80%) till 9th day, while under treated sets, the maximum germination reached till 3rd, 5th or 7th day. Under the treatment of RE, the maximum germination recorded
(100%) with 20% concentration till 7<sup>th</sup> day and with 40 and 60% concentrations on 3<sup>rd</sup> day. With 80% of RE, the maximum germination was 80%

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(non-significant difference with control), but till 3<sup>rd</sup> day as compared to control (9<sup>th</sup> day), while 100% of RE showed 80% germination till 5<sup>th</sup> day. The higher concentrations (120 to 200%) showed significant reduction in G%.

Under SE treatment, the early and maximum germination (100%) was recorded with 20% till 3<sup>rd</sup> day. Other concentrations i.e. 40 and 60% of SE showed 90 G% and 80% of SE showed 80 G% (non-significant) till 3<sup>rd</sup> day, as compared to control (9<sup>th</sup> day). Higher concentrations of SE, from100 to 200% reduced the G% significantly, over control (Table-A; Plate No.-1). Under LE treatment, only with 20% concentration, G% reached upto 100% till 3<sup>rd</sup> day, while other concentrations were either non significant or significantly reduced G%, but the maximum germination reached till 5<sup>th</sup> day. Under FE treatment also, 20% showed 100% germination till 3rd day, as compared to control (80% G% till 9<sup>th</sup> day). Other concentrations i.e. 40% showed 90% germination till 3<sup>rd</sup> day, 60 to 160% of FE showed reduction in the germination and maximum till 3<sup>rd</sup> day, while 200% of FE showed only 20% germination till 5<sup>th</sup> day, (Table-A; Plate No.-2)).

Water Addition

After the initiation of germination the growing seedlings required the addition of water from 5<sup>th</sup> day with different treatments (Table-B). In control sets, water was added every alternate day (7<sup>th</sup>, 9<sup>th</sup>, 11<sup>th</sup>) and in all 26 ml of water is added. Under RE treatment, there was a significant reduction (50%) in the water addition with 20 to 60%, while 80 and 100% of RE showed 46.15%
reduction in water addition. The higher concentrations (120 to 200%) of RE, also showed remarkable reduction in water addition, but followed by reduction in G% (Table A). Under SE treatment, the addition of water started from 5th day due to early maximum germination, as compared to control (7th day). With 20 to 60% of SE, water was added twice i.e. on 5th and 9th day only, while with 80 and 100% of SE water was added on 5th, 7th, and 9th day. With higher concentrations of SE, water addition was reduced with reduction in the germination. Overall, with 20% of SE 53.84% reduction and with 40 and 60% of SE, 50% reduction in water addition was reported, still the G% in these concentrations was higher than control (Table A and B).

Under LE treatment with 20%, 120% and 140% concentrations, water was first added on 5th day. Almost, with all the concentrations of LE water was added either 3 or 4 turns i.e. 7th to 11th or 5th to 11th day (every alternate day). Overall, 20, 40 and 60% of LE showed 53.84 and 61.53% water reduction followed with increase germination till 3rd day (Table A and B). Other concentrations (80 to 200%) showed reduction in water addition (50.00% to 69.23%) with gradual reduction in G%. Under FE treatment, the water addition started at 5th day with almost all the concentrations, except 80 and 200% of FE. The lowest concentration (20%) of FE showed 38.46% reduction in water requirement (100% G%), while 40 and 60% of FE showed 53.84% reduction in water (90 and 70%G%). Higher concentrations from 80 to 200% also showed 50 to 73.07% reduction in water followed with gradual reduction in G% (70 to 20%).
Other Seed Germination Parameters

Other parameters recorded on seed germination showed enhancement with lower concentrations of PE (Table-C and D)

a) Germination Rate Index (GRI)

The data recorded on GRI also showed increase with the concentrations, i.e., 20 to 120% of RE and FE, 20 to 140% of SE, 20 to 100% of LE (Table-C). The higher concentration showed non-significant increase or reduction in GRI. The maximum GRI, i.e., 72.33% was noticed with 40 to 80 of RE, 20% of SE, over control. Among LE treatment, 53.60% GRI was recorded with 20% of LE. Under FE treatment, 20% concentration gave 2nd maximum GRI (64.55%), among all the treatments of PE, over control. The GRI followed the increasing pattern i.e. RE>SE> FE> LE.

b) Speed of Germination (SpG)

SpG was increased with all the concentrations of RE, except 200%, while 100 to 160% showed non-significant increase (Table-C). The range of increase under RE treatment was 26.13 to 278.40% and the maximum was noticed with 40 and 80% of RE. Under SE treatment, almost all the concentrations, except 140 & 200% showed increased SpG and the maximum was recorded with 20% SE (278.40 %), while with increasing concentrations, it was reduced but higher than that of the control up to 120%. LE also increased SpG, only up to 100%, while it was reduced with 120 to 200% of LE over control. The maximum SpG was also recorded with 20% of LE (278.40%) over control. Under FE treatment, SpG was raised with 20 to 100% concentrations
and the maximum was 278.40% with 20% of FE, over control. Similarly, 120 to 200% showed either non-significant increase or reduction in SpG.

c) Coefficient of Velocity of Germination (CVG)

The observations made on CVG showed that it was increased with lower concentration of PE, while higher concentrations i.e. 160 and 200% of RE, LE, and FE, either showed non-significant change or reduction (Table-C). On the other hand, 100 to 200% of SE also showed non-significant increase or reduction. The maximum CVG (i.e. 130%) was

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recorded with 40 and 60% of RE, 20 and 40% of SE, while 20% of LE showed 120% CVG. FE treatment did not showed much variation in CVG (i.e. 120%) with lower concentrations (20 to 100%), while higher concentrations were non-significant.

d) Emergence Index (EI)

The data collected on EI showed that it was increased from 1.50 to 3.75 fold with different concentrations of PE (Fig.-A). Under RE treatment, the maximum EI was recorded with 40% and it was reduced gradually from 60 to 140% of RE, but higher than that of control, while 160 and 200% of RE showed non-significant change. The SE treatment showed increased EI from 2.17 to 3.00 fold with different concentrations and the maximum was noticed with 20% of SE. Similarly, 160 and 200% of SE were non-significant. LE treatment also showed an increase in EI with 20 to 120% concentrations and the optimum concentration was 40% (2.63 fold increase). Under FE treatment, the increase in EI was 1.60 to 1.97 fold recorded with only lower
concentrations (20, 40 and 60%) and the optimum was 40% of FE. Overall, the range of increase showed the pattern with different extracts as RE>SE>LE>FE.

e) Relative Seed Germination (RSG)

The calculation made on RSG showed that it was increased with 20 to 140% of RE, 20 to 60% of SE, 20 to 100% of LE, 20 to 80% of FE (Table-D). The maximum RSG recorded with 20 to 60% of RE and SE (150). 160 and 200% of RE, 140 and 200% of SE, 120 to 200% of LE and 100 to 200% of FE showed significant reduction in RSG.

Results

f) Relative root elongation (RRE)

The data on RRE showed that all the concentrations of LE and lower concentrations of RE were more effective than SE and FE, over control (Table-D). 20 to 140% of RE increased the RRE value and the range of increase was 53.11% to 133.01% with different concentrations and the optimum concentration was 60% of RE. Under SE treatment, 20 and 40% concentrations showed similar RRE as that of control, while 60 to 200% showed significant enhancement in RRE and the best reported with 60% of LE (341.33%), over control. All the concentrations of FE were either non-significant or significantly reduced it.

Seedling Growth Germination Parameters

a) Seedling Growth (SG)
The observation revealed that lower concentrations of all the PE treatments, significantly enhanced the SG (cm) parameter (Table-D), while 140 to 200% of RE and LE, 120 to 200% of SE, and 100 to 200% of FE, either significantly reduced or non- significantly changed the SG. Under RE treatment, the range of increase in SG value varies from 8.36% to 95.28% upto 120% of RE and the maximum increase was reported in 60% of RE. SE treatment showed the increase in SG value from 32.15% to 82.21%, while the optimum concentration was 80% of SE, over control. The different concentrations of LE showed significant increase with 20 to 120% and the maximum was recorded with 80% of LE (137.19%), over control. Under FE treatment, the increase in SG was recorded with only 20 to 80% and the optimum was 60% of FE (82.10%), over control. Overall, LE showed the best increase among all the treatments for SG.

Results

a) Seedling Root Length

The data recorded in root length of seedlings (cm) showed invariable increase i.e. from 1.69% to 27.11% only, over control (Table-E). Under RE treatment, the maximum increase recorded with 120% concentration i.e. 20.33%, over control. SE treatment showed the maximum enhancement with 20% concentration, i.e., 27.11%, over control. On the other hand, LE treatment showed non-significant increase with 20 to 100% concentrations, while 120 and 140% of LE significantly increase the length (18.64%), over control. The FE treatment showed significant enhancement upto 20.33% with 120% of FE, over control. Overall, the increase in root length
was marginal and SE showed the best increase among all the treatments of PE. The higher concentrations (160 and 200%) were all together inhibitory for all the treatments.

c) Seedling Shoot Length

The observation recorded on shoot length of seedlings (cm) shown that the concentrations up to 140% were promotory to increase the length (Table-E). Among all the treatments, SE showed best increase, over control. Under RE treatment, 80% increases the shoot length maximum by 30.23%, over control (8.6 cm). Under SE treatment, the increase in shoot length varies from 24.41% to 39.53%, over control and the maximum was with 100% of SE and LE treatments showed the maximum increase with 120% (31.39%), over control. On other hand, 20 and 40% of FE showed maximum increase in shoot length (33.72%), over control. Among all the treatments, 160 and 200% were inhibitory for shoot length.

d) Fresh Weight of Seedling

The data collected on fresh weight of seedlings (g) showed significant enhancement with all the concentrations of PE, except 160 and 200%, which showed either non-significant increase or reduction, as compared to control (Table-E). Under RE treatment, the range of increase was 4.37 to 18.24% with different concentrations and the maximum was recorded with 120% of RE, over control. SE showed the maximum increase in fresh weight with 40% i.e. 42.25% over control, and 20, 120, 140% of SE also showed the increase, which was similar i.e. 9.48%, over control. Under LE treatment, the lower concentrations (20 to 60%) significantly reduced the fresh weight,
while 80 to 140% showed the increase from 16.78 to 75.18%, over control. The maximum increase was recorded with 120% of LE. Among all the treatments, FE showed remarkable increase and with 80% of FE showed the maximum (82.48%), over control.

e) Dry Weight of Seedlings

The observations on dry weight of seedlings (g) showed that the remarkable dry matter accumulation in the seedlings was with only few concentrations of PE and the rest of the concentrations either significantly reduced or non-significantly affected the dry weight (Table-E). Under RE treatment, 20 to 100% were effective and the increase ranges between 7.5 to 37.5%, over control. The most effective concentration among RE was 40%, also followed by significant increase in fresh weight. SE treatment showed that all the concentrations were either ineffective or non-significantly change the dry weight. Under LE treatment only 80, 100 and 120% were effective to increase dry weight, while the maximum increase was 25.0% with 100 and 120% of LE, over control. On the other hand,

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the only concentration among FE treatments was 60%, which significantly enhance the dry weight up to 47.5%, over control.

f) Vigour Index (VI)

The observation revealed that the data collected on seed germination parameters provide significant change in VI, with lower concentrations of PE, while higher concentrations (from 80% or above) showed either non-significant change or significant reduction in VI (Fig.-B). The lower concentrations, i.e., 20, 40, and 60% of RE, enhanced the VI by 25.00, 47.32, and 28.57%,
respectively over control. Under SE treatment also, 20 to 80% increased the VI and the optimum was 60% of SE, which increased the VI by 42.85%, over control. On the other hand, only 40% of LE showed significant increase in VI (20.53%), over control, while other concentrations were either inhibitory or showed non significant change. Under FE treatment, only 40 and 60% concentrations promoted VI i.e. by 28.57%, over control, rest of the concentrations were inhibitory or non significant. Overall, increasing pattern of VI under different treatments of PE showed as RE>SE>FE>LE.

Section B

Growth and Productivity

The observation taken to study the effect of four different parts of Parthenium extracts on growth and productivity of cowpea, at three stages of plant growth and developments i.e. preflowering, flowering and post flowering (25, 45, 65 DAS, respectively). The results are presented in Tables 1 to 20, Figures 1 to 4 and Plate No. 3 to 8.

Results

A. Morphological Parameters on Growth

a) Root Length

The data collected on root length of cowpea under the treatment of different parts of Parthenium extracts showed an increase with all the concentrations, except 140 % of RE, SE and FE, at all the three stages, 80 to 140% of LE at 65 DAS, over control (Table-1). The increase in root length varies from 2.56% to 22.50% at 25 DAS, 9.45 % to 25.81 % at 45 DAS and 4.30 to 14.39% at 65
DAS, under different concentrations of RE. The maximum increase recorded with 40% of RE i.e., 22.55%, 25.81% and 14.39% at 25, 45 and 65 DAS, respectively over control.

Under the treatment of SE, the root length is increased variably from 8.14 to 18.63% at 25 DAS, 12.44 to 39.10% at 45 DAS and 9.62 to 23.42% at 65 DAS, under different concentrations. The maximum increase recorded with 40% SE at all the three stages (25, 45, and 65 DAS). The higher concentrations (120 and 140%) of SE, initially at 25 DAS showed non-significant change and reduction, while at 45 and 65 DAS, only 40% showed non-significant change.

Under the treatment of LE, the significant increase was recorded with all the concentrations, except 120 and 140% at 45, 65 DAS, which showed non-significant change. The maximum % increase recorded with 20% LE i.e. 19.71, 20.36 and 15.17% at 25, 45 and 65 DAS, respectively over control. Similarly, all the concentrations of FE increased root length, except 120% at 45 DAS and 140% at all the three stages of growth. The maximum increase recorded with 80% of LE i.e. 21.86% and 19.24% at 25 and 65 DAS, respectively, while 40% FE showed maximum increase i.e. 33.48% at 45 DAS. Overall, the increase in root length was more between flowering to post-flowering than pre-

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flowering to flowering. The leaf extract showed comparatively less increase as compared to other extracts of Parthenium parts. The increasing pattern of root length under different extracts followed 40% of SE > 80% of FE > 40% of RE > 20% of LE.

b) Shoot Length

The observation taken on shoot length revealed that it was considerably increased with different concentrations of extracts from 0.83 to 27.27%, over control. The shoot length increase was
more from preflowering to flowering than flowering to post flowering (Table-2). At 25 DAS, different PE showed increase from 6.40 to 27.27% with different concentrations. 40% of RE and SE showed maximum enhancement in shoot length i.e. 27.27% and 26.64%, respectively over control (Plate No.-3 and 4). Comparatively, at 25 DAS, the enhancement in shoot length by different PE followed RE>SE>FE>LE. At 45 DAS, the enhancement in shoot length over control was less than that at preflowering stage. Overall, the increase in shoot length was ranged between 0.83 to 18.38%, with different concentrations of PE. The maximum increase recorded with 40% RE and SE i.e. 12.44% and 18.38%, respectively, while 20% LE and FE i.e. 5.16% and 7.45%, respectively over control. At 65 DAS, the overall increase in shoot length showed 14.46 to 25.01% over control, but the higher concentrations also showed significant enhancement as compared to 45 DAS. The maximum enhancement at 65 DAS recorded with 80% of RE and FE i.e. 23.00% ad 21.41%, respectively, while 40% of SE and 20% of LE, i.e., 25.01 % and 17.89%, respectively over control. Overall, 40% of RE at preflowering stage and 40% of SE at 45 DAS and 65 DAS proved to be optimal concentration for enhancing the shoot length.

Results

The cumulative doses, i.e., treatments given at 20, 40, and 60 DAS showed that the enhancement in shoot length recorded at 65 DAS under different concentrations of PE. The higher concentrations i.e. 140% of RE at 45 DAS, 120% of LE and FE at 45 DAS, 140% of LE and FE at 25 DAS and 45 DAS showed non-significant increase in shoot length. Overall, SE showed significant increase with all the concentrations at all the three stages of growth.

c) Number of Laterals
The observations on number of laterals per plant revealed that at 25 DAS, no lateral branching was recorded. At 45 DAS, number of laterals increased with all the concentrations, except 120 and 140% of SE and LE, while 20, 40, 120 and 140% of LE. At 65 DAS, 140 % of RE and SE, 120 and 140% of LE and FE also showed non-significant change in the number of laterals. Overall, the increase in number of laterals recorded i.e. 9.65 to 39.77% with different concentrations of PE, over control at 45 DAS, while at 65 DAS, 0.97% to 45.28% increase was recorded. The maximum increase observed at 45 DAS, with 40% of RE and FE i.e. 24.85 % and 36.26%, respectively and with 20% SE and 80% LE i.e. 39.77% and 23.39%, respectively over control. The observations at 65 DAS showed the maximum increase with 80 % RE, 40% SE, 80% LE and 40% FE (32.45%, 29.05%, 37.29% and 45.28%, respectively), over control. The cumulative effect of three treatments is visible with 80% of RE and LE, 40% LE and FE at 65 DAS. The increasing pattern of number of laterals under different extracts followed 40% of FE> 80% of LE> 80% of RE> 40% of SE (Table-3).

Results

d) Number of Leaves

The observation recorded on the number of leaves showed little variation at initial stage of growth, i.e., 25 DAS and only 1.2 to 1.3 fold increase was noticed with different concentrations of PE (Table-4). The maximum increase is observed with 40% RE and SE, 20% LE and 80% FE at 25 DAS. At 45 DAS, about 1.2 to 1.4 fold increase in number of leaves was recorded, with different concentrations. While, higher concentration i.e. 140% of SE, LE and FE showed non-significant change. Almost similar observations were recorded at 65 DAS. The increase in
number of leaves was less in control from 25 to 45 DAS than 45 to 65 DAS, while it is vice-versa with different concentrations of PE. 20% of FE showed cumulative effects of three treatments at 65 DAS and the increase is almost two fold, between 45 to 65 DAS. The increasing pattern of number of leaves under different extracts followed 80% of SE> 40% of RE> 80% of FE> 20% of LE.

e) Fresh Weight of Leaves

The observation revealed that all the concentrations of PE significantly increased the fresh weight of leaves/plant (g), except 140% of RE, SE, and LE at 25 DAS, 120% SE and FE at 25 DAS, 140% LE and FE at 45 DAS and 80, 120 and 140% of FE at 65 DAS, (Table-5). Overall, 1.21% to 48.58% increase in fresh weight of leaves is recorded at 25 DAS, 3.13 to 47.91% at 45 DAS and 2.22% to 22.81% at 65 DAS, with different concentrations of PE. The maximum increased was observed with 40% of RE and SE (48.58% and 28.82%, respectively), 20% LE (9.3%) and 80% FE (15.39%), over control at 25 DAS. At 45 DAS, the maximum increase was noticed with 80% RE and SE (47.91% and 52.74%, respectively), 20% LE (34.86%), and 40% of FE (31.46%), over control. At 65 DAS also, 80% of RE and SE showed maximum enhancement i.e. 22.73% and 22.81%, respectively over control. While, 40% of LE showed only 13.89% and 20% of FE showed 12.58% increase in fresh wt. of leaves, at 65 DAS, over control. In untreated controls, the fresh weight accumulation in leaves was more between 45 to 65 DAS than 25 to 45 DAS, while in treated plants vice-versa is the accumulation in fresh weight. The maximum cumulative effect was observed with 80% of SE, as compared to the other treatments at 65 DAS, with three consecutive doses. The
stimulatory effect of different parts of Parthenium extracts is in the increasing manner of 80% of SE> 80% RE> 40% LE> 20% FE.

f) Dry Weight of Leaves

The observations on dry weight of leaves/plant (g) showed 2.41 % to 49.37 % increase at 25 DAS, 3.28 to 56.64% increase at 45 DAS, while 3.06 to 52.00% increase at 65 DAS (Table-6). The maximum increase in dry weight was recorded with 40 % of RE (49.37%), 40 % of SE (37.26 %), 40 % of LE (8.99 %) and 80% of FE (12.73%), over control, at 25 DAS. On the other hand, 80% of RE showed maximum enhancement i.e. 47.94% at 45 DAS and 23.53% at 65 DAS, 40% of SE also showed maximum enhancement at 45 DAS (56.64%) and 65 DAS (52.00%), over control. Similarly, 40% of LE and FE also showed maximum enhancement at 45 and 65 DAS among different concentrations. Overall, 40% of SE showed the best cumulative effect after three doses given at 20, 40 and 60 DAS, among all the treatments of PE. In untreated controls, the dry weight accumulation was more between 45 to 65 DAS than 25 to 45 DAS, while under different treatments the dry weight accumulation is more between 25 to 45 DAS (Pre-flowering to flowering stage) than 45 to 65 DAS (flowering to post-flowering stage), except 120 and 140% of SE and 140% of FE.

Results

The comparative effect of all the concentrations of PE followed the manner of effect as 40% SE>80% RE> 40% FE> 40% LE.

g) Leaf Area

The observations revealed that almost all the concentrations significantly increased the leaf area (cm²/plant), except 140% of RE, LE and FE, at all the three stages of growth, 120% of RE, LE,
at 45 and 65 DAS, 120% of SE at 45 DAS (Fig.-1), which either showed non-significant change or reduction in leaf area. 80% of SE at 45 DAS and 80% of LE at 45 and 65 DAS also showed non-significant change, while 20 and 40% of FE slightly increased the leaf area non-significantly. The increase in leaf area varies from 2.04 to 75.45% at 25 DAS, 1.23 to 36.51% at 45 DAS and 2.25 to 37.77% at 65 DAS with different concentrations of PE. The maximum increased under RE treatment recorded with 40% i.e. 23.89% 26.16% and 12.35% at 25, 45, and 65 DAS, respectively over control. Under SE treatment, the maximum increase in leaf area was observed with 40% i.e. 75.45% and 36.51% at 25 and 45 DAS, respectively, while with 80% i.e. 37.37% at 65 DAS over control.

On the other hand, 20% of LE showed maximum increase i.e. only 15.09, 21.71 and 7.98% at 25, 45, 65 DAS, respectively over control. The FE treatment showed maximum increase with 80% i.e. 25.92, 20.23, and 26.84% at 25, 45 and 65 DAS, respectively over control. Over all, SE showed the best increase among all the treatments and LE showed the least increase. The increasing pattern in the leaf area followed as SE>FE>RE>LE, as cumulative effect at 65 DAS, under the treatments (Fig.-1). Overall, the increase in leaf area is more pronounced between 45 to 65 DAS than 25 to 45 DAS.

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**Results**

h) Fresh Weight of Plant

The observation taken on the fresh weight of plant (g) i.e. stem and root showed significant enhancement with all the concentrations of RE, at all the three stages of growth, except 80, 120 and 140% at 25 DAS (Table-7) The maximum increase in fresh weight of plant under RE treatment recorded with 40% (22.01), at 25 DAS with 80% (23.22 %) at 45 DAS and with 140%
(22.05%) at 65 DAS, over control. On the other hand, 40% of SE showed optimum increase i.e. 56.46% and 22.27% at 25 and 45 DAS, respectively, While 80% of SE showed optimum increase 20.34% at 65 DAS, over control.

The LE and FE showed significant increase in fresh weight at 25 and 45 DAS, but at 65 DAS either non-significant increase or reduction was observed. The optimum concentrations of LE, for increasing fresh weight of plant were 80% (22.01%) at 25 DAS, 40% (8.12% and 2.22%) at 45 and 65 DAS. Similarly, with FE the optimum concentrations were 40% at 25 DAS and 20% at 45 DAS. Overall, the maximum enhancement in fresh weight was recorded with 40% of SE, (56.46%) at 25 DAS, 20% of FE, (26.21%) at 45 DAS and 140% of RE (22.05%) at 65 DAS (Table-7). The increase in fresh weight of plant was more from 25 to 45 DAS than 45 to 65 DAS in both treated and untreated plants. The cumulative effect of three doses is clearly visible under all the treatments, RE and SE were promotory, while LE and FE were inhibitory.

**i) Dry Weight of Plant**

The observation on dry weight of plants (g) showed significant enhancement with all the concentrations of RE, except 140% at 25 and 65 DAS. Under SE treatment, significant enhancement was observed with lower concentrations at 25 and 65 DAS, while higher concentrations showed the reduction. On the other hand, LE and FE showed enhancement in dry weight at 25 DAS only, while at 45 and 65 DAS, all the concentrations were inhibitory (Table-8). Among all the treatments of PE, 20% of RE showed maximum increase i.e. 116.08 % over control at 25 DAS. 45 and 65 DAS, the maximum increase, was recorded with 80% of RE i.e. 23.22 and 15.43%, respectively over control. Overall, the dry matter accumulation in the plant
was observed more between 25 to 45 DAS than 45 to 65 DAS in both treated and untreated plants. As compared to control, more dry matter accumulation was observed with treated plants between 25 to 45 DAS, but vice-versa is the case from 45 to 65 DAS. The increasing pattern of dry weight of plant showed RE>SE>FE>LE.

j) Total Biomass

The data revealed that total biomass (mg/plant) on dry weight basis increased significantly under lower concentrations of RE and SE, at all the three stages of growth (Fig.-2). LE and FE showed enhancement in total biomass, only at 25 DAS with lower concentrations, while all the concentrations of these at 45 and 65 DAS were either inhibitory or non-significant. The range of increase was 5.39 to 64.62% at 25 DAS, 0.72 to 26.61% at 45 DAS and 0.15 to 17.09% at 65 DAS, under different concentrations of PE. The maximum increase was recorded with 40% of SE (64.62%), and other optimum concentrations were 40% of RE (56.92%) increase, 80% of LE (31.54% increase) and 20% of FE (13.85% increase) against control. At 45 and 65 DAS, the maximum increase recorded with 80% of RE i.e. 27.61% and 17.09%, respectively over control. Overall, the increase in total biomass was more between 25 to 45 DAS than 45 to 65 DAS, in both control and treated plants.

Results

A. Productivity

a) Floral Initiation

The data recorded on the opening of first flower showed that all the treatments of PE initiated early flowering, except higher concentrations (Table-9). Under the treatment of RE, the flowers
are initiated early by about two to five days and five days early initiation was observed in 40% of RE. 20 and 40% of SE also showed about 4 days early initiation of flowers. On the other hand, LE showed three days early flowering with 40 and 80% concentrations. The FE also showed early flowering by two days with lower concentrations and 20% of FE was the optimum.

b) Number of Flowers

The observations revealed that number of flowers increased with all the concentrations of PE, except 120 and 140% of SE, LE and FE (Table-9; Plate No.-5 and 6). Overall, 7.62 to 36.73% significant increase was recorded with different concentrations of PE. Among RE treatment, 40% showed maximum increase i.e. 36.73%, over control. On the other hand, 40% of SE, LE and FE were also optimum concentration to increase the number of flowers by 25.62%, 12.33% and 25.97%, respectively over control. The increase in number of flowers under all the PE treatments showed the pattern RE>SE>FE>LE.

c) Number of Pods

The data recorded on number of pods/plant, till maturity showed that pods are increased significantly with all the concentrations of PE (Table-10). The increase ranges between 1.3 to 1.8 fold with different concentrations. The maximum number of pods noticed with 40% of RE (21.66), 80 % of SE (23.07), 40% of LE (20.89), and 40% of FE (22.23), against control (13.00). Overall, 40% of the entire PE is optimum for increasing the number of pods, while higher concentrations (120 and 140%) showed about 1.3 to 1.5 fold increase. The increase in number of pods with different treatments of PE followed the pattern SE>FE>RE>LE.
d) Length of Pods

The data on average length of pods (cm) revealed that it is increased with all the concentrations, except 140% of RE, 120 and 140% of FE and all the concentrations of LE (Table-10; Plate No. 7 and 8). The increase in length varies from 1.94 to 40.16% with different concentrations of PE. The maximum increase observed with 40% of RE (20.57%), 20% of SE (40.16%), 40% of LE (13.77%), and 40% of FE (18.27%), over control. Overall, stem extract proved to be the best among all the treatments. The pattern of increase in length of pods shown by different treatments was SE > RE > FE > LE.

e) Number of Seeds/ Pods

The observation showed a little, but significant variations in number of seeds/pod under different concentrations of PE, except 120 and 140% (Table-11). The increase in number of seeds varies from 3.3% to 18.6% with different concentrations of PE, over control. The optimum concentrations among the treatment were 40% of RE and SE (13.24 % and 18.62%, respectively), 20% LE and FE (12.97% and 9.44%, respectively), over control. Overall, SE proved to be the best for increasing number of seeds/pod. The increasing pattern followed as SE > RE > LE > FE.

f) Weight of 100 Seeds
The weight of 100 seeds (g) showed almost similar pattern as that of number of seeds/pod, except that the 40% concentrations among the entire PE were optimum for the increase (Table-11). The increase varies from 1.53 to 39.00%, over control and the optimum concentrations among the treatment were 40% of RE, SE, LE, and FE (39.00, 36.87, 17.84 and 16.31 %, respectively), over control. Overall, the increasing pattern of 100 seeds weight observed as RE>SE>LE>FE.

g) Seed Yield / Plant

The observations revealed that all the concentrations of PE significantly increased the seed yield/plant, except 120 and 140% of RE, 80, 140% of LE and 140% of FE (Fig.-3). The significant increase varies from 3.53 to 41.02 % with different concentrations of PE, over control. The optimum concentration among the treatment was 40% of RE, SE, LE and FE for increasing the seed yield / plant (25.82%, 41.02%, 6.41 %, and 12.81%, respectively) over control. The increase in yield followed the pattern as SE>RE>FE>LE.

h) Yield

The total yield (q/ha) of cowpea varied greatly under the effect of different PE. The RE and SE significantly enhanced the yield, except 120 and 140%, while all the concentrations of LE showed non-significant increase (Fig.-4). On the other hand, FE with lower concentrations (20 and 40%) increased the yield. The maximum increase recorded with 40% of SE i.e.33.42%, over control. Other optimum concentrations were 40% of RE and FE (25.77% and 12.78%, respectively), over control.
Section - C

Nodulation and Root knot Nematode Studies

I-Nodulation

The observations taken on nodulation parameters (number, size, colour, fresh and dry weights of nodules) and root knot nematode (number of active eggs, J2 stage, galls) and nodule verses nematodes were presented in Tables 12 to 17; Plate No. 9 to 12.

a) Number of Nodules

The effect of different concentrations of PE, on the number of nodules was clearly visible at initial stage of growth i.e. 25 DAS, but later on at 45 and 65 DAS, the lower concentrations significantly increase the number of nodules, while higher concentrations were either inhibitory or significantly reduced the number (Table-12). At initial two stages i.e. pre-flowering and flowering, all the nodules were healthy, while at 65 DAS (post-flowering) senesced nodules were also recorded. At 25 DAS, the increase in healthy nodules varies from 1.21 to 42.32% with different concentrations of PE. The maximum increase recorded with 20% of RE (37.46%), 40% of SE (42.32%), 40% of LE (3.46%) and 40% of FE (24.47%), over control. At 45 DAS, the increase varies from 1.01 to 31.64% with different concentrations and maximum increase in healthy nodules was reported with 40% of RE and SE i.e. 11.07% and 31.64 %, respectively over control.

The LE recorded the maximum increase i.e. only 1.88% with 40%, over control at 45 DAS. On the other hand, all the concentrations of FE significantly reduced the nodules at 45 DAS. At 65 DAS, 20 and 40% of RE non-significantly increased the healthy nodules,
while higher concentrations significantly reduced it. The healthy nodules were significantly increased with 20 and 40 % of SE and 80% of FE at 65 DAS, while other concentrations of SE, LE and FE either showed non-significant change or significantly reduced it. On the other hand, a significant reduction in senesced nodules was recorded at 65 DAS, over control, except 20 and 40% of RE, 20% of SE and 140% of LE (Plate No.-9 and 10).

b) Size of Nodules

The observations taken on minimum and maximum size (mm) of nodule at three stages of growth (25, 45 and 65 DAS) are present in Table-13. The data revealed that at initial stage of growth, there is a little variation in the minimum size of nodules, the lower concentrations 20 and 40% of all the PE showed no difference, but higher concentrations showed the minimum size i.e. 0.90 to 0.94 mm, as compared to control (1.10 mm). The maximum size of nodules at 25 DAS, recorded with 20% of RE i.e. 2.89 mm, as compared to control (2.33 mm). Other optimum concentrations for maximum size of nodules were 20% of SE (2.80 mm), 40% of LE (2.89 mm) and 80% of FE (2.59 mm). The higher concentrations of RE (80 and 120%), SE (140%), LE (140%) and FE (120 and 140%) showed insignificant change in the maximum size of nodules.

At 45 DAS, the higher concentrations of the entire PE showed new nodule development and the minimum size was 0.90 to 1.00 mm, while lower concentrations showed 1.10 mm nodules, as compared to control (1.2mm). At this stage, the maximum size i.e. 3.78 mm (30.80%) of nodules recorded, with 20% of FE, as compared to control (2.89 mm). Other optimum concentrations were 40% of RE (3.56 mm), 80% of SE (3.49 mm), 20 and 40% of LE (3.66 mm), as compared to control (Table 13).
Results

At 65 DAS, the cumulative effect of 20% of the entire PE is observed. The minimum size i.e. 0.94 to 1.0 mm of nodules recorded with higher concentrations of PE i.e. 120 and 140%, respectively, as compared to control (1.11 mm), while 20% and 40% RE showed minimum size (1.33 mm), 20 to 80% SE showed 1.16 to 1.33 mm; 20 to 80% of LE showed 1.26 to 1.33 mm and 20 to 80% FE showed 1.24 to 1.33 mm, minimum size of the nodules. The maximum size of nodules at 65 DAS noticed with 20% of RE (4.83 mm), SE (4.89mm), LE (4.66mm) and FE (4.50 mm), as compared to control (4.11mm). Overall, at post-flowering stage, all the concentrations increased, the size of nodules significantly, except 140% of the entire PE and 120% of LE (Table-13).

c) Colour of Nodules

The colour of nodules showed lots of variations from white to brown grey, at three different stages of growth (Table-14). White is the initiation and brown grey is senescent stage, when nodules were almost decayed. At initial stage i.e. pre-flowering (25 DAS) almost the nodules were white or light pink, while 20% of RE, 20 to 80% of SE and 40, 80% of LE and FE showed pink colour of nodules. At 45 DAS, the control plant showed pink and reddish brown nodules, while the lower concentrations of the entire PE showed white, pink and dark pink nodules. The higher concentrations at 45 DAS also showed reddish brown nodules leading to greying. At post flowering stage (65 DAS), in control plants most of the nodules were brownish grey and few nodules were pink and dark-pink, while under the treatments 20 and 40% of the entire PE showed most of the nodules pink, dark pink, reddish brown and very less nodules brownish grey.
The higher concentrations (80 to 140%) showed few white nodules (new) and also more brownish grey nodules.

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**Results**

d) *Fresh Weight of Nodules*

The observations revealed that the all the concentrations of PE were promotory for fresh weight (mg) of nodules/plant, except 140% of RE, SE, 120 and 140% of LE, at all the three stages of growth (Table-15). On the other hand, all the concentrations of FE at 25 DAS were either non significant or inhibitory, 80 to 140% at 45 DAS, and 120 and 140% at 65 DAS were also inhibitory. The maximum increase in fresh weight of nodules recorded with 40% of SE i.e. 36.39% at 25 DAS, 52.01% at 45 DAS and 87.47% at 65 DAS, over control, Among all the treatments, other optimum concentrations were 20% of RE (28.28%), 20% of LE (14.03%), 20% of FE (5.77%) at 25 DAS, over control.

At 45 DAS, optimum concentrations to increase fresh weight were 40% of RE (47.30%), 80% of LE (18.01%), and 40% of FE (3.65%), over control. At post flowering stage (65 DAS), the cumulative effect of 40% of RE (42.83% increase), 40% SE (87.47% increase), 40% of LE (46.93% increase) and 20% of FE (33.71% increase) was clearly observed, over control. In control plants, the increase in fresh weight was recorded from 25 to 45 DAS, similar with the treated sets, but the reduction in weight under the treated sets is quite less from 45 to 65 DAS, as compared to control. Overall, good number of nodules was observed till post flowering stage. The increasing pattern for fresh weight of nodules with PE was observed as SE > RE > LE > FE (Table- 15).

e) *Dry Weight of Nodules*
The dry weight of nodules also showed almost similar pattern as that of fresh weight. The increase is dry weight was less between 25 to 45 DAS, as compared to the reduction in dry weight from 45 to 65 DAS, in control plant as well as 20% of RE, SE and FE, 80% of RE and 40% of LE (Table-16). On the other hand, other treatments of PE showed more increase in dry weight from 25 to 45 DAS, as compared to the reduction from 45 to 65 DAS (which was less). The range of increase in dry weight was recorded from 12.50 to 57.50% at 25 DAS, 9.82 to 48.39% at 45 DAS and 6.85 to 83.01% at 65 DAS, over control. Overall, 40% of SE and RE showed better cumulative effect than other concentrations. The increasing pattern for cumulative concentrations observed as SE>RE> LE>FE.

II-Root Knot Nematode studies

a) Number of Active eggs

The observation revealed that the effect of PE, when directly added to the isolated nematodes in aqueous solutions were inhibitory to the number of eggs and active adult nematodes (Table-17 a; Plate No.-11 a, b and 12 a, b). After 24 hrs of the treatment the higher concentrations, 80 to 140% were inhibitory, than lower concentrations. Overall, FE showed maximum reduction in the number of eggs (48.16 to 80.39%) from 20 to 140%.

After 24 hrs of direct treatment, the number of eggs significantly reduced from 16.54% to 59.77%, with different concentrations of PE and higher concentrations reduced more as compared to lower concentrations (Plate No.-11 a, 12 a). After 48 hrs of treatment (Plate No.-11 b, 12 b, the number of eggs is commonly reduced in control (without Parthenium extract) as well as in treated sets. At this stage, the significant % reduction varies from 24.18 to 80.39%, with
different PE and from 40 to 120% showed more reduction than lower concentration (20%). After 72 hrs of treatment again, the number of eggs increased in control as well as treated sets. After 72 hrs, the lower concentrations of RE (20 and 40%) showed more recurrence of active eggs, as compared to control, while 80% of RE showed less recurrence and reduced 42.73%, over control. Under SE treatment, 20 to 120% showed less recurrence and reduced 19.78 to 62.56%, over control. Under LE treatment, 20 and 40%, showed less recurrence and reduced 30.48 and 27.32%, respectively over control. Under FE treatment, only 20% showed less recurrence and reduced the active eggs (44.76%), over control (Table-17 a).

b) Number of J₂ Active Nematodes

On other hand, solution from control plants showed three active nematodes / unit area of magnification after 24 hrs of collections, while under the treatments, number of active J₂ nematodes were reduced to 1 or nil (Plate No.-11 and 12 a). After 48 hrs of treatment, no active J₂ nematodes were observed with >40% concentrations (Plate No.-11 and 12 b), but after 72 hrs of treatment 1 or 2 active nematodes were observed with 20 to 80% of RE and SE. Under LE and FE treatments, no active nematodes were observed even after 72 hrs of the treatment (Table-17 b).

c) Number of Root Knots Galls

The data collected on number of galls developed on the root surface showed remarkable reduction with different concentrations of PE, when sprayed at three stages of growth (Table-
The galls present on untreated plants were 13 at 25 DAS and subsequently increased with the increasing age of plant i.e. 17 at 45 DAS and 24 at 65 DAS. On the other hand, all the treatments of PE reduced the galls by 30.76% to 100%. The RE treatment showed maximum reduction in galls (100%) with 140%, while 20 to 120% showed 30.76 to 84.61% reduction at 25 DAS. At 45 DAS, with 2 doses of RE subsequently increased the reduction, while 58.82 to 100% with increasing concentrations from 20 to 140% of RE. Similarly, at 65 DAS, the reduction was more prominent i.e.. 75% to 100%. With three consecutive doses (20, 40, 60 DAS), 80 to 140% of RE showed no galls on the root system.

Under SE treatment, almost similar observations were made as that with RE, but 80% of SE with 2 doses (20 and 40 DAS) did not allowed the formation of galls (Table-17c). The lower concentrations (20 and 40%) of SE significantly reduced the galls by 62.50% and 83.33%, respectively over control. 120 and 140% of SE completely checked the formation of galls at vegetative stage (25 DAS) and didn’t allowed the formation of galls even at later stages of growth.

LE treatment also showed significant reduction in the number of galls by 53.84 % to 100%, at three different stages of growth. 20% of LE reduced the galls by 53.83% at 25 DAS, 64.70 % at 45 DAS and 79.16% at 65 DAS, while 40% SE showed 69.23% reduction at 25 DAS, 88.23 % reduction at 45 DAS (with two doses) and completely checked the formation of galls at 65 DAS (three doses). The higher concentrations (80 to 140%) LE completely checked the gall formation at 45 DAS.

Under FE treatment, the maximum reduction in the galls was observed at 25 DAS, with all the concentrations from 53.84 to 100%. With different concentrations (20 to 140%), over control,
while 40 to 140% of FE completely checked the formation of galls at 45 DAS (two doses). At 65 DAS, 20% of FE showed 50% reduction in galls, while other concentrations didn’t allowed the development of galls on the roots.

d) Nodule verses RKN

The observations recorded on number of nodules as well as number of galls developed on the root system showed that the higher concentrations of RE i.e. 80 to 140 % with three consecutive doses significantly reduced both the parameters. With lower concentrations (20 and 40%) of RE on one hand, significantly reduced the galls by 58.82 and 70.58%, over control and on the other hand, significantly increased the number of nodules upto 45 DAS by 4.09% and 11.07%, respectively over control (Table- 12 and 17c; Plate No.-9). At 65 DAS, 20 and 40% of RE reduced number of galls as well as showed non-significant change on the number of nodules (Table-12 and 17c, Plate No.-10).

Under SE treatment, the number of galls significantly reduced with all the concentrations at all the three stages of growth, while the significant increase in number of nodules was recorded with all the concentrations, only at 25 DAS, 20 to 80% at 45 DAS, 20 and 40% at 65 DAS. Three consecutive doses of SE with 80 to 140%, completely checked the development of galls, but also reduced the number of healthy nodules at 65 DAS (Table - 12 and 17 c; Plate No.-9 and 10).

LE treatment, with all the concentrations were more deleterious for root knots development as well as didn’t increased the number of nodules significantly, even the lowest concentration (20%), which reduced the galls 79.16%, also non-significantly affect the number of nodules at all the three stages of growth. 40% of LE reduced the galls 69.23% at 25 DAS, 88.23% at 45 DAS
and 100% at 65 DAS, while the number of nodules under LE treatment, reduced only 3.46% at 25 DAS, 1.88% at 45 DAS, and non-significantly changed at 65 DAS.

Under FE treatment, at 25 DAS the reduction in galls from 53.84% to 100% with increasing concentrations from 20 to 140% of FE, while lower concentrations (20 and 40%) of FE slightly increased the number of nodules by 19.08% and 24.47%, respectively over control (Table-12 and 17c). At 45 DAS, 20% of FE reduced the galls by 52.94% and other concentrations (40 to 140%) by 100%, while all the concentrations of FE

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significantly reduced the number of nodules (Plate No.-9). At 65 DAS, 20% of FE reduced the galls by 50% and other concentrations completely checked the development of galls, while only 80% of FE increased the nodules by 13.64% and rest of the concentrations either reduced the number of nodules or showed non-significant change (Plate No.-10)

Section D

Morphological Stomatal studies

a) Stomatal Frequency

The observation recorded on stomatal frequency at three different stages of plant growth, are presented in Tables-18a, b, c. The results indicated that stomatal frequency was increased significantly with lower concentrations of PE, while with 80% or >80% the frequency was reduced or non-significantly changed, on both adaxial and abaxial surfaces.

I Treatment (20 DAS)

(i) On Adaxial Surface
When the treatment has been given at 20 DAS, the untreated controls didn’t show any variation in the stomatal frequency upto three subsequent days (21, 22, 23 DAS), while different concentrations of PE showed increase under RE treatment on 21 DAS. The stomatal frequency significantly increased by 32.69 to 65.45% with different concentrations, except 140% RE and the maximum with 40%. On subsequent days 22 and 23 DAS, the increase recorded as 65.91 and 66.46% with 40% of RE, over control (Table-18 a). After 3rd DAT (23 DAS), the higher concentrations 80, 120 and 140% showed non-significant change in stomatal frequency.

**Results**

Under SE treatment, the stomatal frequency was slightly higher than RE treatment and it was increased gradually on three subsequent days (21st, 22nd, 23rd) after I treatment, with all the concentrations, except 120 and 140% on 21 and 22 DAS. The maximum increase recorded with 40% of SE i.e. 66.13% on 21st day followed with 66.40% on 22 DAS and 66.53% at 23rd day. LE treatment showed less increase in stomatal frequency than SE and RE with slightly variable results. At 21st day, the maximum frequency was observed with 40% of LE (56.67%), but it was sharply increased on 22 DAS (57.41%), followed by 23 DAS (58.66%), over control. FE treatment also showed less increase in stomatal frequency than SE, RE, and LE and initially 1st DAT (21 DAS), the maximum increase was 35.63% with 40%, while on 22 DAS, the increase was 47.94% with 20% of FE and then it was slightly reduced to 44.91% on 23 DAS with 20% of FE. The reduction in stomatal frequency followed the pattern FE<LE<RE<SE.

(ii) **On Abaxial Surface**

As compared to adaxial surface, the stomatal frequency was more on abaxial surface. In untreated controls the frequency didn’t vary in subsequent three DAT (21, 22, 23 DAS), while
under PE treatment, it was increased significantly (Table-18 a). Under RE treatment, all the concentrations, except 140% on 21 DAS, increased the stomatal frequency and the maximum frequency was observed with 80% of RE i.e.16.20% on 21 DAS, 28.27% on 22 DAS and 32.36% on 23 DAS. With SE treatment, the stomatal frequency was increased significantly over control and more than RE treatment .The maximum frequency was observed with 80% of SE, on three subsequent DAT i.e.17.44% at 21 DAS, 29.50% at 22 DAS and 32.78% at 23 DAS. LE treatment also showed increase in stomatal frequency over control, but less than that of RE and SE. The maximum increase observed with 40%

Results

of LE i.e.11.74% at 21 DAS, with 80% i.e.23.74% at 22 DAS and followed by 28.17% at 23 DAS. On the other hand, with FE treatment initially 20% of FE showed maximum increase in stomatal frequency at 21 DAS (5.72%), while at 22 DAS, 80% of FE showed maximum i.e. 14.05% followed by 18.29% with 80% at 23 DAS. The reduction in stomatal frequency followed the pattern FE<LE<RE<SE.

II Treatment (40 DAS)

(i) On Adaxial Surface

The observations revealed that a stomatal frequency at flowering stage, in control plants remain same when study on three consecutive days, i.e., 41, 42, and 43 DAS, while under different concentrations of PE, it has been increased slightly (Table-18 b). Under RE treatment, the stomatal frequency was significantly increased, except 120 and 140% at 43 DAS (3rd DAT) and the maximum frequency was observed with 80% i.e. 12.76% at 41 DAS, 13.35% at 42 DAS and 13.97% at 43 DAS. Under SE treatment, the lower concentrations were stimulatory to increase
the frequency, while 120 and 140% were non significant, and the maximum stomatal frequency was observed with 80% of SE, which was almost similar on two consecutive days i.e. 13.38% at 41 DAS and 13.56% at 42 DAS, but slightly increased i.e. 15.71% at 43 DAS.

Under LE treatment, the II treatment showed that lower concentration increased the stomatal frequency, but less than that of RE and SE and the maximum increase recorded with 40% at 42 DAS (9.50%) and 43 DAS (10.64), while at 41 DAS, it was only 6.36%, over control. The 80% of LE, although showed maximum increase at 41 DAS (9.40%), but on subsequent days i.e. 42 and 43 DAS it remained almost same (9.42% and 9.00%).

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FE treatment also increased the stomatal frequency, but the significant increase was recorded only with 20% and on 42 DAS it was less than 41 DAS. The maximum enhancement in frequency was 4.61% at 43 DAS. Other concentrations of FE showed no significant change on stomatal frequency. The cumulative effect of two doses shows the pattern of maximum increase as 80% of SE>80% of RE>40% of LE> 20% of FE. The reduction in stomatal frequency followed the pattern FE<LE<RE<SE.

(ii) On Abaxial Surface

On the abaxial surface, the stomatal frequency was more at flowering stage than pre-flowering stage, about 21 stomata in control sets, while in treated sets, it was less than control (Table-18 b). Under RE treatment, the maximum increase in stomatal frequency was recorded in 40% i.e. 8.14% at 41 DAS, 9.19% at 42 DAS and negligibly increased at 43 DAS (9.65%), over control.
The higher concentrations (80 to 140%) initially showed non-significant difference at 41 DAS, slight reduction at 42 DAS, while significant increase at 43 DAS, from 10.40 to 11.43% with increasing concentrations, over control. Almost similar observations were recorded with SE treatment and maximum increase was with 40% of SE i.e. 8.83% at 41 DAS, 10.33% at 42 DAS and 10.99% at 43, DAS. The higher concentrations (80 to 140%) of SE showed slight increase at 41 DAS (2.12 to 5.52%), while reduction was noticed at 42 DAS and again enhancement at 43 DAS.

LE treatment showed that the stomatal frequency was initially increased with 20 and 40% (2.77 and 6.93%, respectively) at 41 DAS. From 41 to 42 DAS, it was increased by only 3.51 to 4.61% or non-significantly changes on the 3rd DAT (43 DAS), again the frequency

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Results

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was increased and the maximum was recorded with 20% of LE (6.65%), over control. Under FE treatment, not much variation was observed at 41 DAS, while stomatal frequency was reduced with 20 and 40% of FE at 42 DAS (0.94 and 1.00%) respectively, over control. Higher concentrations of FE (80 to 140%) showed non-significant effect on stomatal frequency. The cumulative effect of two doses showed the increasing pattern as 40% RE > 80% SE > 20% LE > 40% FE. The reduction in stomatal frequency followed the pattern FE < LE < SE < RE.

III Treatment (60 DAS)

(i) On Adaxial Surface

At 3rd stage of growth, i.e., post-flowering, the stomatal frequency was slightly increased, from 2.75% to 7.48%, with different concentrations of PE (Table-18 c). Under the RE treatment, the lower concentrations showed an increase in stomatal frequency on 1st DAT after III treatment (61
DAS), which remained unchanged on 2\textsuperscript{nd} DAT (62 DAS), while slightly increased on 3\textsuperscript{rd} DAT (63 DAS). The maximum increase was recorded with 80% of RE i.e. 5.35% at 61 DAS, 5.28% at 62 DAS and 6.90% at 63 DAS, over control. SE treatments also showed similar observations with all the concentrations, except 140% increased the stomatal frequency and 80% showed the maximum increase i.e. 6.75% at 61 DAS, 6.11% at 62 DAS, and 7.48% at 63 DAS. The lower concentrations (20 and 40%) of SE also showed 4.91 and 6.95% increase, over control.

Under LE treatment, 80% showed maximum increase at 61 DAS (6.40%), while at 62 and 63 DAS stomatal frequency was reduced. On 62 and 63 DAS, maximum increase was recorded with 40% of LE (2.83 % and 5.28%, respectively), over control. 120 and 140% of LE non-significantly changes the stomatal frequency. On the other hand, all the concentrations of FE showed non-significant or significant reduction at all the three DAT (61, 62, 63 DAS). The reduction % varies from 4.65 to 8.84% at 63 DAS, with different concentrations of FE. The increase in frequency followed the pattern 80% of SE> 80% of RE> 40% of LE

(ii) On Abaxial Surface

The stomatal frequency was more on abaxial surface than adaxial surface in control as well as in treated sets (Table-18 c). The 3\textsuperscript{rd} treatment at 60 DAS showed that under different treatment the stomatal frequency increased from 2.87 to 5.08% at 61 DAS, 2.83 to 5.87% at 62 DAS, 1.69 to 5.19% at 63 DAS over control. Under RE treatment, maximum frequency was observed with 80% i.e. 3.81% at 61 DAS, 5.31% at 62 DAS, and 4.34% at 63 DAS, while higher concentrations (120 and 140%) were non-significantly for increasing the frequency. SE treatment also showed the similar observation to increase the stomatal frequency by 5.19% at 63 DAS,
while higher concentrations were either inhibitory or non-significant at 61 DAS, while slightly promotory at 62 and 63 DAS.

Under LE treatment, the effect was slightly variable that 20, 40 and 80% concentrations initially increased the stomatal frequency at 61 DAS, while reduction upto 63 DAS, but more than that of control. The maximum increase was recorded with 40% of LE i.e. 2.73% over control at 63 DAS. 120 and 140% of LE showed non-significant change in stomatal frequency. On the other hand, all the concentrations of FE either showed non-significant or significant reduction in stomatal frequency, on three subsequent days of III treatment (Table-18 c). The increasing pattern with cumulative doses observed as 80% of SE> 80% of RE> 40% of LE.

Results

**Cumulative Effect of PE**

The observations on stomatal frequency clearly showed that the cumulative effect of two and three doses of PE, at three stages of plant growth (pre-flowering to flowering, two doses; flowering to post-flowering, three doses), significantly reduced the stomata, as compared to control (Table-18 a, b, c). The results indicated more increase in stomatal frequency with two cumulative doses than three cumulative doses i.e. between 23 to 43 DAS than 43 to 63 DAS. With the different concentrations of PE, the increase with cumulative doses was quite less than control. On the adaxial surface, 20% of RE, SE, LE and FE proved to be more effective for increasing minimum number of stomata, while on abaxial surface, 80% of RE, LE and FE were effective with two cumulative doses, but with three cumulative doses the effective concentrations were 40% of RE, 20% of SE, LE, and 40 % of FE.

b) **Stomatal Index (SI)**
The observation recorded on stomatal index at three different stages of plant growth, are presented in Tables-19 a, b, c.

I Treatment **(20 DAS)**

(i) *On Adaxial Surface*

The stomatal index was calculated on three consecutive days after I treatment i.e. on 21, 22 and 23 DAS, and the result present in Table-19 a showed that there was a very little variation in SI, as compared to control. On the whole, 40% of RE, 20% of SE, LE and FE showed maximum SI (7.00, 3.43, 3.43, 3.43%, respectively), over control on 3\textsuperscript{rd} DAT (23 DAS). The higher concentrations (120 and 140%) of PE were non-significant to increase the SI.

(ii) *On Abaxial Surface*

On the adaxial surface, initially at 21 DAS the SI was increased with RE than slightly reduced at 22 DAS, and again increased at 23 DAS, while with other treatments, like SE initially SI was increased and gradually raised up up to 23 DAS. Under LE treatment, the SI was reduced, except 20 and 140% at 21 DAS, than slightly increased at 22 and again reduced at 23 DAS, over control. FE showed significant or non-significant reduction in SI at 21 DAS than slightly increased at 22 and 23 DAS (Table-19 a). Overall, maximum SI recorded with 120% of RE (9.69%) 80% of SE (10.37%), 20 to 80% of LE (3.48%, same) and 20% of FE (6.27%) at 23\textsuperscript{rd} DAS, over control.

II Treatment **(40 DAS)**

(i) *On Adaxial Surface*
Stomatal index was increased with almost all the concentrations of PE, except 140% of SE and LE, as compared to control (Table-19 b). Under RE treatment, one day after II treatment (41DAS), the lower concentrations i.e. 20% initially increased the SI, which gradually became more at 42 DAS, then again reduced. 40% of RE showed gradual reduction in SI, from 41 to 43 DAS (2.27 to 1.59%). The higher concentrations (80 and 120%) slightly reduced the SI from 41 to 42 DAS, than increased from 42 to 43 DAS. Similarly, SE treatment also increased the SI, except higher concentrations, which showed non-significant change, over control. Under LE treatment, 20% initially increased the SI, then became non-significantly different at 42 and 43 DAS, while other concentration (40 to 140%) were inhibitory or non-significant at 41 DAS and gradually increase the SI till 43 DAS, except 140%. On the other hand, FE initially showed no change with 20 to 80% concentrations at 41 DAS, then gradually increased at 42 and 43 DAS, while 120 and 140% showed an increase in SI, which gradually reduced on subsequent days. Overall, on 3rd DAT, the optimum concentrations were 80% of RE (2.02%), 40% of SE (0.43%), 40% of LE and FE (2.02 and 2.39%, respectively), over control.

(ii) On Abaxial Surface

On the abaxial surface, the lower concentration, i.e., 20% of RE and FE showed non-significant effect on SI, while other concentrations increased it. 40% of RE increased SI slightly from 41 to 42 DAS than reduced it on 43 DAS. 80 and 120% of RE increased the SI significantly at 43 DAS, while 140% at 41 DAS and then it was increased. 40 and 80% of SE showed significant
increase at 41 DAS, which was gradually reduced till 43 DAS. On the other hand, almost all the concentrations of LE gradually increased the SI from 41 to 43 DAS and more than that of control. FE treatment showed variable effect on SI i.e. all the concentrations of FE significantly increase SI on 41 DAS, while 20 and 40% of FE reduced the SI on 42 DAS than increased on 43 DAS, while 80% of FE showed no change in SI on 42 and 43 DAS, which was almost similar on all the three days (minimum increase in SI, over control). The higher concentrations 120 and 140% of FE showed gradual increase in SI from 41 to 42 DAS (Table-19 b). The minimum % increase in SI was observed in 20% of RE, SE and LE (1.02, 2.14 and 2.94%, respectively) and 80% of FE (2.89%), over control.

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**Results**

**III Treatment (60 DAS)**

(i) *On Adaxial Surface*

The observations recorded after III treatment (60 DAS) showed that there was a little variation in SI on 61 DAS and almost similar to SI of II treatment (40DAS). Under different treatments, SI was slightly increased with lower concentrations of PE, while higher concentrations were almost inhibitory (Table-19 c). The lower concentrations of RE i.e. 20, 40 and 80% increase the SI by 0.42, 7.38 and 1.56%, respectively over control on 63 DAS. SE treatment also showed increase in SI from 61 to 63 DAS with 20 to 80% concentrations, while 20 and 40% of SE slightly reduced SI from 61 to 62 DAS, over control. The higher concentrations (120 and 140%) of RE increased the SI, while SE reduced the SI or showed non-significant change. On the other hand,
20 and 40% of LE initially reduced the SI on 61 DAS, then increase the SI upto 63 DAS, but the maximum increase was only 1.02%, over control. FE treatment showed a non-significant increase with 20 and 40% on 61 DAS, and then only 20% of FE increases the SI on 62 DAS, while all the concentrations significantly reduced it on 3rd DAT (63DAS).

(ii) On Abaxial Surface

Number of stomata as shown by the stomatal Index was more on the lower surface (abaxial) as compared to adaxial. All the treatments of PE showed variable change in SI initially on 61 DAS, except 120 and 140% of RE (Table-19 c).

Under RE treatment, 20, 40 and 80% concentrations reduced the SI from 61 to 62 DAS and again increased it on 63 DAS. Among lower concentrations, 20% of RE showed minimum increase in SI i.e. 5.17% over control. SE treatment also showed increase in SI, initially at

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61 DAS, then gradually reduced upto 63 DAS, which was less than that of control. Under LE treatment, SI was more with lower concentrations (20 to 80%), while higher concentrations (120 and 140%) showed the reduction on 63 DAS. On the other hand, all the concentrations of FE showed gradual increase in SI from 61 to 63 DAS, over control. Overall, the increase was not remarkable with all the concentrations of FE, over control, but the reduction was observed with all the concentrations of SE and LE.

Anatomical Studies on Stomata

The anatomical studies on stomata, like variations in guard cells (GCs), subsidiary cells (SCs) and stomatal aperture are presented in Plate No.13-48.
a) Effect of Root Extracts

I Treatment (20 DAS)

In cowpea, the stomata are paracytic type with two bean shaped GCs, slit like aperture and five SCs of almost equal size on both the surfaces, but several variations, like unequal GCs, number of SCs, and stomatal aperture was observed under different treatments of PE. Basically, the stomata in control plants were bigger on adaxial surface than abaxial surface. The effect of I treatment of RE was given at 20 DAS and the observations were taken on three subsequent days i.e. 21, 22 and 23 DAS. The findings are presented in Plate No.- 13 to 15.

On 21 DAS, the effect of RE treatment was clearly visible with the slight variations in the shape and size of GCs (Plate No.-13 A3, A4, A6 on adaxial surface; B2, B3, B4, B6 on abaxial surface). These variations were continuous on 22 and 23 DAS also, which was visible by Plate No.- 14 A3, A4, A6, B2, B3, B5, B6; 15 A2 to A6, B2 to B6. Similarly, the

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effect of RE on the number and size of SCs was also visible. The higher concentrations i.e. 120 and 140% showed four SCs on 21, 22 and 23 DAS (Plate No.-13 A5, A6, B5, B6; 14 A5, A6, B5, B6; 15 A6, B5, B6), on adaxial as well as abaxial surface. 120% of RE, on adaxial surface (Plate No.-15 A5) and 40% of RE on abaxial surface (Plate No.-14 B3), showed three SCs. The stomatal aperture appeared to be small under the treatments, except 120% on 23 DAS (Plate No.-15 A5), 20% on 22 and 23 DAS (Plate No.-14 B2; 15 B2), 140% on 22 and 23 DAS (Plate No.-14 B6; 15 B6), 40% of RE on 22 and 23 DAS (Plate No.-14 B6; 15 B6) of RE on abaxial surface showed abnormality in one GC, which appeared to be almost half of the second GC (Plate No.-15 B3).
II Treatment (40 DAS)
The II treatment was given at 40 DAS (flowering stage) showed variation in stomata and SCs. The structural variations are presented in Plate No.- 16 to 18. The higher concentrations showed much variation i.e. unequal size of GCs (Plate No.-16 A2 to A6, B2 to B6; 17A3, A6, B2 to B6), on adaxial as well as abaxial surface. These structural changes in GCs slightly overcome on 43 DAS (Plate No.-18).

The effect of RE treatment, was also observed on the number of SCs, which were reduced to four with 140% of RE on 41 and 42 DAS (Plate No-16 A6; 17 A6), on adaxial surface, 120% of RE on both the surfaces (Plate No.-16- B5; 17 A5, B5; 18 A5, B5). The size of stomata with guard cells was also reduced with higher concentrations as compared to control (Plate No.- 16, 17, 18-B5). 80 and 140% of RE also showed one GC almost half size of the other GC on 41 DAS (Plate No.-16 B4, B6).

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III Treatment (60 DAS)
The III treatment was given at the stage of post-flowering (60 DAS) and the effect of three doses were clearly visible (Plate No.-19, 20 and 21). All the concentrations of RE, significantly reduced the number of SCs and the size of GCs varies. The unequal size of GCs was observed just after the III treatment on 61 DAS, which was continued to 62 DAS and slightly reduced on 63 DAS. On the other hand, 80, 120 and 140% of RE clearly showed variation in the size of GCs (Plate No.-19 A4 to A6, B4, B6; 20 A5, A6, B4 to B6; 21 A5, A6, B4, B6) from 61 to 63 DAS, on the both the surfaces. The number of SCs were observed three (Plate No.-19 B2; 21 A4) four (Plate No.- 19 A2 to A6, B3 to B6; 20 A2 to A6, B2 to B6; 21 A2, A3, B2 to B5). The stomatal
aperture appear to be more open under the treatment or almost similar that of the control (Plate No.-19 A4; 20 B3; 21 A2, A4, A6, B5, B6). 80% of RE showed a common SC between two stomata (Plate No.-20 B4).

**b) Effect of Stem Extract**

**I Treatment (20 DAS)**
The observations recorded on 1<sup>st</sup>, 2<sup>nd</sup>, 3<sup>rd</sup> DAT (21,22,23 DAS, respectively) showed that all the concentrations, except 20% of SE invariably affect the GCs, SCs and stomatal aperture. 20 and 40% of SE showed slight enlargement in SCs, but the size of GCs and the number of SCs almost remain same. The higher concentrations, 80 to 140% showed reduction in number of SC i.e. four on adaxial surface (Plate No.-22 A4, A5, A6; 23 and 24 A5, A6) and 120 and 140% on abaxial surface i.e. 3 or 4 (Plate No.-22, 23, 24, B5, B6), while 80% concentration showed unequal or uneven division in SCs, on 22 and 23 DAS,

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on adaxial surface (Plate No.-23 and 24 A4) and on abaxial surface almost normal, but slightly enlarged SCs was observed (Plate No.-22-24 B4). 120% of SE also showed variation in GC i.e. one of the GC either wavy or reduced in size (Plate No.-22-24 A5, B5), 40% and 80% of SE on abaxial surface showed unequal size of GCs, almost one of the GC about 2/3 of other (Plate No.-22-24 B3, B4). 140% of SE, also showed unequal GCs, on adaxial surface and on abaxial the GC initially wavy, then reduced in size into unequal GC. (Plate No.-22- 24 A6, B6). In control, the stomatal aperture is slit like and open from 21 to 23 DAS (Plate No.-22-24 A1, B1), while under treatment, the aperture was small and slightly open. The smallest aperture recorded with 40% on the abaxial surface (Plate No.-22-24 B3).
II Treatment (40 DAS)
The effect of spray treatment at 40 DAS clearly showed the effect on number and size of SCs, size of GCs and stomatal aperture (Plate No.- 25-27) on 41 to 43 DAS. Initially, at 41 DAS, the number of SCs remained to be five, except 40 and 140% on adaxial surface (Plate No.- 25 A3, A6) and 80% on abaxial surface (Plate No.-25 B4), although the size and shape of the SCs vary. 20, 40 and 80% of SE showed enlargement in one or two SCs on both the surfaces (Plate No.- 25 A2, A3, A4, B3, B4). 120% also showed unequal division in SCs, resulted in six SCs (Plate No.- 25 B5), at 41 DAS.

In subsequent days i.e. 42 and 43 DAS, almost similar observations were made and less number and large size SCs were observed with 40, 80, 120, and 140% concentrations (Plate No.-26 A3, A5, A6, B4; 27 A2, A3, A6, B3 to B6). Unequal size of GCs were observed with almost all the concentrations, on both the surfaces in three subsequent days and one of the GCs either reduced in size or appear to be wavy (Plate No.- 25 A4, A5, B2, B4, B6; 26 A2, A3, A6, B5, B6; 27 A2, A5, A6, B6). On the other hand, 40% of SE showed butterfly wing shaped GC on 43 DAS on adaxial surface (Plate No.-27 A3). The stomatal aperture was slightly reduced, but remained open under the treatment.

III Treatment (60 DAS)
After III treatment, at 60 DAS (post- flowering stage) the normal size and shape of SCs and GCs appear to be slightly smaller than flowering stage (40DAS). The three subsequent doses showed the cumulative effect on SCs, GCs, and aperture. Just one day after treatment (61DAS), the number of SCs reduced to three or four, on both the surfaces (Plate No.- 28 29 and 30 A3 to A6, B4 to B6) The lower concentrations i.e. 20% of SE showed five SCs with variable sizes (Plate

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A2, A3, A6, B5, B6; 27 A2, A5, A6, B6).
No.-28-30 A2), on adaxial surface, while on abaxial surface it showed four SCs of large size at 61 DAS then number increase to five with two small and three large SCs, on 62 and 63 DAS (Plate No.- 28-30 B2).

On the other hand, the effect on GCs was clearly visible as unequal size of GCs with different concentrations and 120% showed one or both the GCs as wavy (Plate No.- 28-30 A5, B4, B6). The stomatal aperture appeared to be round, with 80% of SE on adaxial surface (Plate No.-28-29 A4), with 20 and 40% on abaxial surface (Plate No.-29-30 B2, B3).

**c) Effect of Leaf Extract**

I Treatment (20 DAS)
After the I treatment at pre-flowering stage (20 DAS), variability in stomatal structure were also observed with LE as the reduction in SCs, size of GCs and stomatal aperture (Plate No-31 to33). The variations that occur on 21 DAS also continued to 23 DAS. Initially after

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1st DAT, the enlargement in SCs was observed on both the surfaces, with all the concentrations of LE, except 40% on adaxial surface (Plate No.-31 A3). The number of SCs were observed four with all the concentrations, except 40 and 80% on adaxial surface (Plate No.-31, A3, A4), in which five SC were observed, similar to that of control. On abaxial surface, with 20% few of the stomata were seen sharing common SC (Plate No.-31 B2). Later on, 2nd DAT and 3rd DAT the number of SCs increased with 20, 40, 80 and 120% on adaxial surface and with 40% on abaxial surface (Plate No.-32, A3, A5, B3; 33 A2, A3, A4, B3).

Similarly, the size of GCs was also more that of control on 22 and 23 DAS (Plate No.-32 and 33). On the other hand, the size and shape of GCs was also varied with all the concentrations of
LE, as compared to control. The higher concentrations showed much variations like wavy GCs or one of the GC very small (Plate No.-31 A4, A6, B2, B3, B4, B6; 32 A3, A4, A6, B2, B3, B5; 33 A2, A3, A4, A6, B6). The size of stomata also reduced almost half or less then half under the treatment, either on 21, 22 and 23 DAS (Plate No.-31 B4; 32 A4, B3, B5, B6, 33-A3, A4, B2, B6). As the size is reduced, the stomatal aperture either appeared as narrow slit like (Plate No.-31, A5, B5; 33 A2, B6) or appeared as small rounded pore (Plate No-31 B3; 32-B6; 33 A4, B2, B4, B5).

II Treatment (40 DAS)
The II treatment given at flowering stage also showed variations in the size and number of SCs, size and structure of GCs, as well as the stomatal aperture (Plate No.-34-36). Just after 1st DAT, the variations were clearly observed in SCs i.e. three to four SCs with enlarge size under 40 to 140% of LE, on both the surfaces (Plate No-34 A3, A5, A6, B2, B3, B5, B6). On 2nd DAT, the variations were almost similar as that of 1st DAT that the reduced number of SCs with all the treatments, except 20% on abaxial surface, showing five SCs as the divisional stage (Plate No-35 B2), while shape of SCs under the treatment was enlarged and irregular, as compared to control. At 3rd DAT, the effect of lower concentrations i.e. 20% was slightly reduced and the number and shape of SCs became normal (Plate No.- 36 A2), while other concentrations, still showed variation in number and size of SCs as three or four, either very big or very small in size (Plate No.-36 A5, A6, B4, B5, B6). Similarly, the GCs also showed variation in size with all the treatments i.e. one of the GC smaller than the other, at all the three subsequent DAT, except 20% on 42 and 43 DAS (Plate No.-35, 36, A2). On the other hand, 80% of LE, on adaxial surface (Plate No.-34-36 A4), 40% and 120%, on adaxial surface at 41 DAS

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and 80%, at 42 and 43 DAS showed one of the GC wavy (Plate No.-34 B3, B5; 35 and 36, B4). Initially the higher concentrations, 120 and 140% showed one of the GC very large with open slit like aperture (Plate No.-34 to 36 B5, B6), while other concentrations showed normal aperture according to the size.

III Treatment (60 DAS)
III treatment given at post-flowering stage, showed the cumulative effect three doses on number of SCs, shape and size of SC, GCs and stomatal structure (Plate No.-37 to 39). The numbers of SCs were four with all the concentrations of LE, except 140% on adaxial surface, which showed three SCs of large size (Plate No.-37 to 39 B6). The size of GCs also vary with treatment as one of the GC became very small and other very large (Plate No.-37 A2 to A6, B3, B5; 38 A4, A6, B2, B5; 39 A2 to A6, B4, B6). Some of the variations in GCs, like wing shape (Plate No.-37 A2, A4, B3; 38 A2, A5; 39 A4, A5) or wavy (Plate No.-37 A5, A6; 38 A2, A4, A5, B6; 39 A3, A6) were also observed. The size and shape of aperture also varies, as the size of stomata small, the aperture became round shaped or small pore like (Plate No-37 A3, A6, B2; 38 A4, A6; 39 B4), as compared to control and other concentrations, showed slit like open aperture.

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d) Effect of Flower Extract

I Treatment (20 DAS)
After 1 treatment of FE, the observation taken on subsequent 1st, 2nd, 3rd DAT, some variations were observed in the shape, size, and number of SCs, size of GCs and stomatal aperture (Plate
Overall, 20% of FE showed slight enlargement in SCs on both the surfaces, which appeared to be under divisional stage on further two days (Plate No.-40 to 42 A2, B2), but stomata appeared to be normal and slightly reduced in GCs. With other concentrations, four SCs were observed with enlarged and variable shapes, like winged, flattened or wavy (Plate No.-40 A5, A6, B4, B5, B6; 41 A3, A6, B4, B5; 42 A2, A5, B3, B5, B6). On the other hand, the size of GCs was also affected with FE treatment as unequal GC was observed with 40, 80, 140% of FE, on adaxial surface at 21 DAS (Plate No.-40 A3, A4, A6), with 20 and 140% on abaxial surface (Plate No.-40 B2 to B6). Similarly, on 22 and 23 DAS, unequal GC were observed with all the concentrations, except 20%, where GCs were smaller than the control, but appeared to be normal (Plate No.-41 and 42, A2, B2). The stomatal aperture appeared to be more opened, but slightly smaller according to the size of GCs. The round shaped aperture observed with 40%, on adaxial surface, at 21 DAS, 80% on abaxial surface at 22 DAS (Plate No.-40 A3; 41 B4). Narrow slit like aperture was observed with 140%, on adaxial at 21 DAS, 40% on abaxial at 22 DAS (Plate No.-40 A6; 41 B2).

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II Treatment (40 DAS)
After II treatment, the observations recorded on 1st, 2nd and 3rd subsequent DAT, showed that the concentrations, from 20 to 80% did not much vary the stomatal structures, while 120 and 140% of FE reduced the number of SCs invariably and enlarged them (Plate No.-43-45 A5, A6, B5, B6). Other concentrations also increased the size of two to three SCs, while other SCs appeared to be normal. Some of the variations were observed in SC, as butterfly wing shaped (Plate No.-43 A3, B5; 44 A2, A6, B2; 45 A3, B4, B5, B6). Some of the SCs appeared to be rounded or elliptical in shape (Plate No.-43 A3, B5). On the other hand, the unequal size of GCs was
observed with almost all the concentrations, except 20% on abaxial surface (Plate No.-43 to 45, B2). With some concentrations, one of the GC became invariably enlarge than the other (Plate No.-43 A2, A3, A6, B4, B5; 44 A2, A3, B5, B6; 45 A2, A3, 5, B4, B5, B6). The effect of FE on stomatal aperture was not remarkable, but it was reduced or increased with the size of GCs.

III Treatment (60 DAS)

After III treatment, the observations taken on subsequent three days 1\textsuperscript{st}, 2\textsuperscript{nd} and 3\textsuperscript{rd} DAT, showed that the FE treatment didn’t affect significantly the stomatal structures, except the enlargement in SCs and unequal size of GCs (Plate No.-46 to 48). 20% of FE showed extra enlargement in SCs and GCs on all the three DAS (Plate No.-46 to 48 A2). The higher concentration (140%) reduced the number of SCs to four and all the four SCs were irregular or wavy (Plate No.-46 to 48 A6, B6). On the other hand, unequal size of GCs observed with almost all the concentrations, except 20% on the abaxial surface (Plate No.-46 to 48, B2) and 140% on the adaxial surface and 80% on the abaxial surface (Plate No.-

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46 A6, B4; 47 B4). The maximum effect of 20% of FE observed as enlarged GCs than control and the effect was continued till 63 DAS (Plate No.-46 to 48 A2). The stomatal aperture didn’t show many variations, except 140% on abaxial surface, where it seems to be almost rounded (Plate No.-46 B6).

Section-E

Biochemical Estimations

The observations recorded on biochemical changes in cowpea viz. Chlorophyll content, Total protein and Lb contents in nodules, Protein and Total Soluble sugar contents in dry seeds have been presented in Figures- 5 to 11.
**a) Chlorophyll Content (Chl)**

(i) Chl ‘a’

The observation revealed that Chl ‘a’ (mg/g fr. wt. of leaves) was increased with lower concentrations (20 to 80%) of PE, at all the three stages of growth. At 25 DAS, from 31.25 to 143.75% increase was recorded in Chl ‘a’ with different concentrations of PE. The optimum concentrations for increasing Chl ‘a’ were 40% of RE (100%), 80% of SE (143.75%), 40% of FE (100%), over control. At 45 DAS, Chl ‘a’ increased from 8.57% to 91.43%, with different concentrations of PE. The optimum concentrations among different extracts were 80% of RE and SE (51.43 and 91.43%, respectively), 40% LE and FE (28.57% and 40.00, respectively), over control. At post-flowering stage (65 DAS), similarly 40% of RE, SE, LE and FE showed optimum increase (27.45, 33.33, 35.29 and 15.69%, respectively), over control. The higher concentrations of the entire PE were either non-significant or reduced the Chl ‘a’. The increase in Chl ‘a’ was more between 25 to 45 DAS than 45 to 65 DAS, in both control and treated plants. The cumulative effect of initial two doses at 20 and 40 DAS were more promotory than cumulative three doses. The cumulative effect of three doses were observed as 40% of SE> RE> LE> FE (Fig.-5).

(ii) Chl ‘b’

The observations revealed that Chl ‘b’ (mg/g fr. wt. of leaves) almost showed the similar pattern as that of Chl ‘a’ (Fig.-6). Overall, all the concentrations of RE, SE, and LE increased the Chl ‘b’ at 25 DAS, while higher concentrations of FE significantly reduced it. With the increasing age i.e. at flowering (45DAS) and post- flowering (65DAS), the higher concentrations of SE, LE,
and FE were inhibitory. Normally, the increasing pattern was more between 25 to 45 than 45 to 65 DAS in control plants, while treated plants with all the concentrations, except 20% of RE, 20 to 80% of SE, 20 and 40% of LE showed more increase in Chl ‘b’ between 45 to 65 DAS than 25 to 45 DAS. On the other hand, under FE treatment, all the concentrations showed more increase between 25 to 45 DAS than 45 to 65 DAS, similar to control. Among all the treatments, RE showed cumulative effect of three treatments, higher than other treatments. The increasing pattern was quite variable with different treatments as 80% of RE followed by 20% of LE, 40% of SE and FE (40.33, 40.68, 33.33, 32.20%, respectively over control). Overall, the treatments showed the promotory effect on Chl ‘b’ as RE> SE> LE> FE.

(i) Total Chl

The observations revealed that total chlorophyll content (mg/g fr. wt. of leaves) was enhanced with all the concentrations of PE, except 140% of SE and LE at 25 DAS. 120 and 140% of RE, SE and LE at 45 DAS, 80 to 140% of FE at 25 and 45 DAS, 120 and 140% of

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SE, LE and FE at 65 DAS (Fig.-7). The maximum increase was recorded with 80% of RE (69.99), SE (100.97%), 20% of LE (61.17), 20% of FE (5.83%), at 25 DAS. Among different treatments, at 65 DAS the optimum concentrations were 80% of RE and SE (30.05% and 27.46%, respectively), 40% of LE and FE (14.51% and 15.03%, respectively), over control. On the other hand, at 65 DAS, a little variation was observed with three cumulative doses and optimum concentrations were 40% of RE, SE and FE (40.61%, 32.75% and 28.38%, respectively) and 20% of LE 36.68%, over control. The increase was more between 25 to 45
DAS than 45 to 65 DAS in control sets. Similar increase was observed with lower concentrations of RE (20%), FE (20 and 40%), while other concentrations of PE showed more increase between 45 to 65 DAS than 25 to 45 DAS. The cumulative effect of three doses was maximum with 40% of RE followed by 40% SE, 20% LE and 40% of FE. Overall, all the concentrations of FE were less promotory for total chlorophyll content. The increasing pattern among different treatments observed as 40% RE > 40% SE > 20% LE > 40% FE.

b) Total Protein Content in Nodules

The data recorded on total protein content (mg/100mg fr.wt.) revealed that it was increased with all the concentrations of PE, except 140% of RE at 45 DAS, 120% of SE at 45 and 65 DAS, 140% of SE and FE, at all the three observations, 120% and 140% of LE at all the three observations, 80% of LE and FE and 120% of FE at 65 DAS (Fig.-8). At 25 DAS, the significant % increase range from 14.29% to 77.55% with different concentrations of PE. The optimum increased in nodular protein among the treatments were, 40% of RE (71.43%), 40% of SE (77.55%), 20% of LE (24.49%) and 20% of FE (32.65%), over control at 25 DAS. At 45 DAS, the significant increase varies from 21.69% to 50.60% with different concentrations, over control. The optimum concentrations, with two doses were

80% of RE (49.40%), 40% of SE (50.60%), 20% of LE (31.33%), and 20% of FE (45.79%), over control. At 65 DAS, with three cumulative doses, the maximum increase recorded with 40% of SE i.e. 35.96%, over control, while other optimum concentrations were 40% of RE (22.23%), 20% of LE and FE (17.98% and 16.85%, respectively) to increase the total protein in nodules, over control. Overall, increase was recorded between 25 and 45 DAS (pre-flowering to
flowering), while protein was reduced from 45 to 65 DAS (flowering to post flowering). The reduction between 45 and 65 DAS was less than that of control, with lower concentrations of PE.

c) Leghaemoglobin Content (Lb) in Nodules

The observations recorded on Lb content (mg/100mg fr.wt) in nodules clearly revealed that lower concentrations were more effective, at all the three stages of growth (Fig.-9). At 25 DAS, 20 and 40% of RE (55.39 and 76.95%, respectively), 20, 40 and 80% of SE (46.74, 94.14, and 32.42%, respectively), 20% of LE and FE (16.79 and 62.50% respectively), significantly increased the Lb content. The optimum concentrations were 40% of RE and SE, 20% of LE and FE, over control. At 45 DAS, similarly the lower concentrations were effective with two cumulative doses (at 20 and 40 DAS). The range of increase varies from 8.22% to 67.51% with different concentrations of PE and the maximum increase recorded with 40% of RE (67.51%), over control. At this stage, 40% and 80% of LE also showed cumulative effect as compared to 25 DAS. At 65 DAS, the range of increase was 7.62 to 144.45% with different concentrations of PE, over control. The optimum concentrations among different treatments were 40% of RE (82.47% increase), 20% of SE (65.79% increase), 40% of LE and FE (144.45 and 70.32%, respectively), over control. Overall, with three cumulative doses at 65 DAS, higher concentrations of RE (80 to 140%), SE (120

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**Results**

and 140%), FE (80 to 140%) either showed non-significant increase or reduction in Lb content, while all the concentrations of LE, except 80% showed significant increase in Lb content (Fig-9). Overall, in all the sets, Lb content was increased from 25 to 45 DAS and reduced from 45 to 65 DAS, but the reduction was less prominent in the treated plant nodules than control.
d) Lb: Protein content

The observation on Lb content, which is the part of total nodular protein, in terms of Lb: protein ratio revealed that it varies with different concentrations over control (Table-20). At 25 DAS, the lower concentrations of RE, i.e., 20 and 40% showed better fraction of Lb i.e. 1: 5.62 and 1:6.18, over control (1:6.38). The lower concentrations of SE i.e. 20% and 40% also showed improvement in Lb content with the total protein i.e. 1:6.12 and 1: 5.83, while LE showed lower fraction of Lb: protein against control. On the other hand, only 20% of FE showed optimum fraction of Lb with protein i.e. 1:5.20.

At 45 DAS, i.e., at flowering stage, the Lb: protein ratio has been increased in control, as well as in lower concentrations of RE and all the concentrations of FE, while SE and LE showed almost marginal change in the ratio against control. The best fraction was observed with FE, but higher concentration (140%) showed reduction in Lb content. In control plants, the Lb: protein ratio observed as 1:3.89, while with 20 and 40% of RE, it showed improvement i.e. 1: 3.43 and 1: 3.39. Under SE treatment, all the concentrations have increased, both Lb and protein contents, except 140% against control, but the ratio of Lb to protein is slightly reduced. Similarly, the lower concentrations of LE (20 to 80%) also showed enhancement in both Lb and protein contents, but Lb: protein ratio was reduced (Tables-8, 9). On the other hand, all the concentrations of FE, except 140% increased the protein content significantly and a marginal reduction was observed in Lb content over control. This has improved the ratio of Lb: protein.
At 65 DAS (post-flowering stage), both protein and Lb contents has been reduced from 45 DAS, still both recorded more with the treatments, over control. The Lb: protein ratio was 1: 10.60 with control, while it has been improved with lower concentrations of RE (20 and 40%), SE (20 to 80%), LE (all the concentrations) and FE (20 to 80%). The optimum concentrations for Lb: protein ratio were 40% of RE (1:6.99), 20% of SE (1:7.40), 40% of LE (1:4.92), and 40% of FE (1:7.06) against control (1: 10.60).

e) Total Protein Content in Seeds

The observations taken on protein content (mg/100mg dry wt.) in seed showed that it was remarkably increased with lower concentrations of all the treatments of PE (Fig.-10). Overall, the range of increase was 2.57% to 26.50% with different concentrations of PE and the maximum was recorded with 40% of RE. Other optimum concentrations were 40% of SE (25.64%), 20% of LE (20.51%) and 20% of FE (21.36%,) over control. The higher concentrations i.e. 140% of RE, 120 and 140% of SE and LE, 80 to 140% of FE either showed non-significant change or reduction in protein content. The pattern of maximum increase with different treatment was 40% RE> 40% SE> 20% FE> 20% LE.

f) Total Soluble Sugar Content in Seeds (TSS)

The observations revealed that TSS (mg/100mg dry wt.) in dry seeds was also enhanced with different concentrations of PE, except 140% of RE and LE, 40 to 140% of SE, over control (Fig.-11). Under the treatment of RE, the range of increase was 8.75 to 23.35% and the maximum with 40% of RE. With SE treatment, the increase was less than RE i.e. 4.37 and 12.40% with 40% and 20%, respectively, over control.
Under LE treatment, all the concentrations increased TSS from 4.37 to 15.32%, over control, which was also less than that of RE. On the other hand, with FE treatment 20 and 40% concentrations increased TSS i.e. 10.94 and 4.37%, respectively over control. The maximum increasing pattern among different treatments of PE showed 40% RE > 20% LE > 20% SE > 20% FE.