4. RESULTS

In the present study, results made on diversity, occurrence, distribution, biochemical characteristics and immunostimulant potential of seaweeds occurring along the coast of Thondi coastal regions(Palk Bay), Ramanathapuram district, Tamilnadu, India are presented as follows.

4.1 Seaweeds -Diversity, occurrence and distribution

Four different localities such as Nambutalai(Station-I),Thondi(Station-II),Muthuramalingapattanam or M R Pattanam(Station-III) and Theerthandathanam(Station-IV) with luxuriant growth of seaweeds were selected for this study.

1. Station-I : Nambutalai is a small coastal town located 2 km south of Thondi (Station-II) and more than 200 country boats are operated for fishing every day.
2. Station-II: Thondi is in the Palk Strait in Ramanathapuram District of Tamil Nadu. It is one of the main fishing stations where more than 250 country and mechanized boats are operated every day for fishing.
3. Station-III: M R Pattanam is located 3 km away from Station II towards north and it is a small coastal village without fishing activities.
4. Station-IV : Theerthandathanam(Station- IV) is a pilgrimage centre for Navagreha worship. This Station is 12 km away from station III towards north where 200 country boats are operating every day for fishing.

A total of 25 species belonging to red, brown and green seaweeds have been recorded in the present investigation made at four localities of Thondi and

adjourning regions. Of these, 9 genera with 16 species are red algae (*Rhodophyta)*, 2 genera with each one species are brown algae (*Phaeophyta*) and 5 genera with 7 species are green algae (*Chlorophyta*). Among the four localities, all 25 species were recorded at Theerthandathanam(Station- IV) as maximum diversity followed by 24 species at Thondi(Station-II), 21 species at M R Pattanam(Station-III) and 17 species at at Nambutalai(Station-I). High species diversity was recorded at Theerthandathanam(Station- IV) and Thondi(Station-II) and M R Pattanam(Station-III) and optimum species diversity was observed at M R Pattnam(Station-III), and less number of species was observed at Nambutalai(Station-I)(Fig.1).

Among the different seasons, species diversity was high and more are less same during monsoon and post-monsoon seasons, whereas in pre-monsoon season less number of species was observed in all the localities. The number of seaweeds was very low during summer season in all the localities studied. In Nambutalai(Station-I),11 red seaweeds such as *Amphiroa fragilissima, Gracilaria corticata* var. *corticata, G. corticata* var. *cylindrica, G. edulis,G. verrucosa,G. foliifera ,Hypnea flagelliformis, H.musciformis, H. valentiae , Spiridia hypnoides* and *Acanthophora spicifera;*and green seaweeds such as *Enteromorpha flexuosa , E. intestinalis ,Ulva lactuca ,Ulva reticulata ,Chaetomorpha linum* and *Cladophora facicularis* were recorded during post-monsoon and monsoon seasons whereas brown seaweeds were not observed in all four seasons. During pre-monsoon, 8 red and 5 green algae were recorded whereas in summer a least diversity of 4 red algae(*Gracilaria corticata* var. *corticata, G. corticata* var. *cylindrica, G. edulis* and *G. verrucosa*) and 4 green

algae(*Enteromorpha flexuosa* , *E. intestinalis* ,*Ulva lactuca* ,*Ulva reticulata* and *Chaetomorpha linum*) were observed(Fig. 2).

At station-II (Thondi), all species of red except *Centroceras clavulatum,* brown and green algae were recorded during post-monsoon and monsoon seasons but no record of brown seaweeds during pre-monsoon and summer was found. Ten species of red seaweeds such as *Amphiroa fragilissima, Jania rubens, Gracilaria corticata* var. *corticata, G. corticata* var. *cylindrica, G. edulis,G. verrucosa, H.musciformis, H. valentiae, Spiridia hypnoides* and *Acanthophora spicifera;*and 5 green seaweeds such as *Enteromorpha flexuosa* , *E. intestinalis* ,*Ulva lactuca,Ulva reticulata* and *Chaetomorpha linum* were recorded during pre-monsoon season. In summer, a low diversity of 5 red algae such as *Gracilaria corticata* var. *corticata, G. corticata* var. *cylindrica, G. edulis,G. verrucosa* and *H. valentiae*) and 6 green algae such as *Enteromorpha flexuosa* , *E. intestinalis* ,*Ulva lactuca,Ulva reticulata, Chaetomorpha linum* and *Cladophora facicularis* were recorded(Fig.2).

All the red seaweeds except two red seaweeds *Champia parvula* and *Centroceras clavulatum,* brown seaweed *Chnoospora implexa* and all green algae except *Caulerpa scalpeliformis* were observed at M R Pattanam(Station-III) during post-monsoon season. As in the post-monsoon season, similar species of seaweeds except *Grateloupia filicina* were observed during the monsoon season. An optimum species diversity of 10 red algae such as *Amphiroa fragilissima, Jania rubens, Gracilaria corticata* var. *corticata, G. corticata* var. *cylindrica, G. edulis,G. verrucosa, H.musciformis, H. valentiae, Spiridia hypnoides* and *Acanthophora spicifera* and 5 green seaweeds such as *Enteromorpha flexuosa* , *E. intestinalis* ,*Ulva lactuca,Ulva reticulata* and *Chaetomorpha linum* were

recorded during the pre-monsoon season. As in the pre-monsoon season, same green algal species were recorded in summer and only less number of 6 red seaweeds such as *Amphiroa fragilissima, Jania rubens, Gracilaria corticata* var. *corticata, G. corticata* var. *cylindrica, G. edulis* and *G. verrucosa* were also observed. No brown algae species were found at M R Pattanam(Fig.2).

All the 25 seaweeds were recorded in station IV(Theerthandathanam) during post-monsoon and summer seasons. But brown algae were not observed during summer and pre-monsoon seasons. Eleven red seaweeds such as *Amphiroa fragilissima, Jania rubens, Gracilaria corticata* var. *corticata, G. corticata* var. *cylindrica, G. edulis, G. verrucosa, Hypnea flagelliformis, H.musciformis, H. valentiae, Spiridia hypnoides* and *Acanthophora spicifera* were recorded during the pre-monsoon season. Less number of 8 red algae such as *Amphiroa fragilissima, Jania rubens, Gracilaria corticata* var. *corticata, G. corticata* var. *cylindrica, G. edulis, G. verrucosa, H. valentiae,* and *Acanthophora spicifera* were recorded during summer whereas 6 green seaweeds such as *Enteromorpha flexuosa , E. intestinalis , Ulva lactuca ,Ulva reticulata, Chaetomorpha linum* and *Cladophora facicularis* were observed during pre-monsoon and summer(Fig. 2).

Species vice distribution of red, brown and green algae is shown in Table 1. Red algae such as *Jania rubens, Gracilaria corticata* var. *corticata, G. edulis, G. verrucosa,* and *Acanthophora spicifera* were found most abundant or abundant in all four stations during post-monsoon and monsoon seasons and abundant during summer and pre-monsoon seasons whereas other red seaweeds such as *Amphiroa fragilissima, G. corticata* var. *cylindrica, Gracilaria cannaliculata, G. foliifera, Hypnea flagelliformis, H.musciformis, H. valentiae,*

Champia parvula, Centroceras clavulatum and *Spiridia hypnoides* were recorded as less abundant or sparse. *Grateloupia filicina* was found as sparse during monsoon and post-monsoon seasons and nil during summer and pre-monsoon seasons. Significantly, agarophytes *Gracilaria corticata* var. *corticata, G. corticata* var. *cylindrica, G. edulis* and *G. verrucossa* were found as most abundant during post-monsoon and monsoon seasons and abundant even during summer and pre-monsoon seasons in all localities. Brown algae *Dictyota dichotoma* and *Chnoospora implexa* occurred sparsely during post-monsoon, pre-monsoon and monsoon seasons at Station-II, III and IV except during the pre-monsoon season of Station-III, summer at all localities and all seasons at Station-I where both algae did not occur. Among the green seaweeds, *Enteromorpha flexuosa , E. intestinalis ,Ulva lactuca* and *Chaetomorpha linum* occurred as most abundant or abundant whereas *Ulva reticulata* and *Cladophora facicularis* were found as less abundant or sparse in all the four stations. *Caulerpa scalpeliformis* was observed most abundantly or abundantly during post-monsoon/ monsoon season and sparsely during pre-monsoon or summer season in Station-II, III and IV and absent during summer season in station II and III and all the seasons of Station-I (Table 1).

4.2 Biochemical Study-Proximate composition of seaweeds

The following results were made on the biochemical characteristics related to growth such as dry weight, ash content, total chlorophyll, phycocyanin, allophycocyanin, phycoerythrin and total lipids from fresh samples(Table 2 and 3) ; and total carbohydrate, total protein, total amino acids, total phenol, water retention capacity (WRC),phycocolloids(agar, carrageenan and alginate) and sulphate(Table 4 and 5) from the crude carbohydrate extracts of frozen sample of

16 red, 2 brown and 7 green seaweeds collected from the Thondi coast during November 2007.

4.2.1. Dry wt.

Dry weights of seaweeds are presented in Table 2. Red alga *Jania rubens* significantly recorded a maximum dry weight of 144.56±3.6 mg g^{-1} fresh wt. among all the seaweeds. There were significant differences in the dry weight recorded within red seaweeds. However, differences in dry weight between agarophytes *Gracilaria corticata* var. *corticata* and *G. corticata* var. *cylindrica*;between *G. edulis* and *G. verrucosa* not significant statistically. Among agarophytes, maximum dry weight of 138.2±2.7 mg g^{-1} fresh wt. was registered in *G. corticata* var. *cylindrica.* Optimum dry weight was recorded in two agarophytes *G. edulis* and *G. verrucosa.* Among the carrageenophytes *Hypnea flagelliformis* and *H.musciformis,* between *H.musciformis* and *H. valentiae* no significant difference in dry weight was recorded. Minimum dry weight of 60.7±5.2 mg g^{-1} fresh wt. was observed in *Champia parvula* among the red algae. Even though a high dry weight of 130.7±6.4 mg g^{-1} fresh wt. was registered in *Chnoospora implexa,* difference was not significant with another brown alga *Dictyota dichotoma.* Among the green algae, maximum dry weight of 90.8±3.7 mg g^{-1} fresh wt. was recorded in *Ulva lactuca* compared to *Ulva reticulata* difference was not significant but with other green algae significant difference was noticed. Minimum dry weight of 42.2±5.7 mg g^{-1} fresh wt. was observed in *Cladophora facicularis* among the three groups of seaweeds.

4.2.2. Ash content

Ash content of seaweeds are presented in Table 2. Like dry weight, ash content also recorded maximum 39.6±4.6 mg g^{-1} dry wt. in red alga *Jania rubens* but difference was not significant with *G. verrucosa* (37.9±2.6 mg g^{-1} dry wt.)

whereas they showed significant difference among other red seaweeds. Minimum of 16.4±2.4 mg g^{-1} dry wt. of ash was recorded in *Acanthophora spicifera* among the red algae but it did not exhibit any significant difference with *Centroceras clavulatum* and *Spiridia hypnoides* and showed difference with the rest of the red algae. High ash content of 17.9 ± 3.8 mg g^{-1} dry wt. was registered in *Dictyota dichotoma* but no significant difference was seen with another brown alga *Chnoospora implexa.* Even though maximum ash content of 18.9 ± 3.7 mg g^{-1} dry wt. was recorded in *Chaetomorpha linum,* differences in ash content with other green algae *Ulva lactuca* and *U. reticulata* were not significant but they showed significant difference with the rest of the green algae. Minimum ash content of 09.4±2.1 mg g^{-1} dry wt. was recorded in green alga *Cladophora facicularis* among the seaweeds studied.

4.2.3. Total lipids

Like ash content, significantly a maximum of 67.23±3.6 mg g^{-1} fresh wt. of total lipid was recorded in *G. verrucosa* among the red seaweeds. Differences in total lipid observed among the red algae were significant but among *G. corticata* var. *cylindrica, G. edulis* and *Champia parvula;* between *Gracilaria corticata* var. *corticata* and *G. foliifera;* between *Hypnea flagelliformis* and *H.musciformis; among Amphiroa fragilissima, Centroceras clavulatum* and *Acanthophora spicifera* differences were not significant. A minimum of 27.42±4.1 mg g^{-1} fresh wt. of total lipid was recorded in red alga *Jania rubens,* among all seaweeds. Significant difference in total lipid was recorded among the two brown algae and high total lipid of 31.18±2.1 mg g^{-1} fresh wt. was recorded in *Chnoospora implexa.* Significantly a maximum total lipid content of 78.76±6.2 mg g^{-1} fresh wt. was recorded in green alga *Caulerpa*

scalpeliformis among all the seaweeds but showed no significant difference with another green alga *Ulva lactuca.* Minimum total lipid content of 37.41±2.7 mg g^{-1} fresh wt. was recorded in green alga *Cladophora facicularis* among the green seaweeds studied(Table 2).

4.2.4. Total Chlorophyll

Total chlorophyll was higher in green seaweeds than red and brown seaweeds whereas accessory pigments were higher in red and brown seaweeds than green seaweeds. Among the accessory pigments, commonly phycoerythrin was high followed by allophycocyanin and phycocyanin in all the three group of seaweeds. Generally, total chlorophyll was high in green seaweeds followed by red and brown seaweeds. Of the 17 red seaweeds, *Gracilaria verrucosa* recorded maximum total chlorophyll of 0.38±0.004 mg g^{-1} fresh wt. but difference was not significant with *Gracilaria corticata* var. *corticata* and *Acanthophora spicifera* and showed significant variation in total chlorophyll with the rest of the red algae. Another agarophyte *G. edulis* showed moderate total chlorophyll content of 0.34±0.003 mg g^{-1} fresh wt. but it did not show any significant total chlorophyll content with *G. corticata* var. *cylindrica* and *G. canaliculata.* Minimum total chlorophyll of 0.07±0.002 mg g^{-1} fresh wt. was observed in *Jania rubens* among the red algae. Less amount of total chlorophyll content of 0.07±0.001 mg g^{-1} fresh wt. and 0.06±0.002 mg g^{-1} fresh wt. was recorded in brown algae *Dictyota dichotoma* and *Chnoospora implexa* ,respectively and the differences did not exhibit significant variation. Among the green algae, maximum total chlorophyll of 1.89±0.64 mg g^{-1} fresh wt. was recorded in *Ulva lactuca* which is the highest among the three groups of seaweeds recorded. Differences in total chlorophyll content among the green

algae were significant. Next to *Ulva* species, high total chlorophyll of 1.77±0.62 mg g^{-1} fresh wt. was recorded in green alga *Chaetomorpha linum.* Low total chlorophyll content of 1.04±0.32 mg g^{-1} fresh wt. was observed in *Cladophora facicularis* among the green seaweeds (Table 3).

4.2.5. Phycocyanin

Generally phycocyanin was higher in red and brown algae than green algae. Among the red algae, high phycocyanin of 0.38±0.002 mg g^{-1} fresh wt. and 0.38±0.004 mg g^{-1} fresh wt. were recorded in *G. edulis* and *G. verrucosa,* respectively and differences among them were not significant. Differences of phycocyanin content among the red algae were not mostly significant. Minimum phycocyanin content of 0.03±.001 mg g^{-1} fresh wt. was observed in *Jania rubens* among the red seaweeds. Compared to some red algae , phycocyanin content of two brown algae was low and differences in the phycocyanin content between *Dictyota dichotoma*(0.31 ± 0.001 mg g^{-1} fresh wt.) and *Chnoospora implexa*(0.29 ± 0.001 mg g^{-1} fresh wt.) were insignificant. Compared to red and brown algae, phycocyanin content was less in green seaweeds. Differences in phycocyanin content were not significant among the green seaweeds (Table 3).

4.2.6. Allophycocyanin

Commonly, allophycocyanin was higher in red and brown algae than green algae. Allophycocyanin content was higher than phycocyanin in all seaweeds. Among the red algae, significantly high allophycocyanin content of 0.51±0.02 mg g^{-1} fresh wt. was recorded in *G. verrucosa* which was the maximum among all the three group of seaweeds investigated. Differences in allophycocyanin content among the red algae were not quite significant.

Minimum allophycocyanin content of 0.17±0.00 mg g^{-1} fresh wt. was observed in *Jania rubens* among the red seaweeds. Compared to some red algae, allophycocyanin content of two brown algae was low and significantly higher allophycocyanin content of 0.26±0.01 mg g^{-1} fresh wt. was recorded in *Dictyota dichotoma* than *Chnoospora implexa* (0.17±0.01 mg g^{-1} fresh wt.). Compared to red and brown algae, allophycocyanin content was less in green seaweeds. Differences in allophycocyanin content were not significant among the green seaweeds (Table 3).

4.2.7. Phycoerythrin

Among the accessory pigments, phycoerythrin content was high followed by allophycocyanin and phycocyanin in all the three groups of seaweeds. Of the red seaweeds, like phycocyanin and allophycocyanin significantly high phycoerythrin content of 0.77±0.04 mg g^{-1} fresh wt. was recorded in *G. verrucosa* which was the maximum among the all three group of seaweeds investigated. Differences in phycoerythrin content among the red algae were not quite significant. Next to *G. verrucosa* , *G. edulis* recorded a high phycoerythrin content of 0.71±0.03 mg g^{-1} fresh wt. among the red, brown and green algae investigated. Among the red seaweeds, minimum phycoerythrin content of 0.35±0.03 mg g^{-1} fresh wt. was observed in *Jania rubens*. Compared to red seaweeds, phycoerythrin content of two brown algae was low and showed significant differences between *Dictyota dichotoma* (0.34±0.02 mg g^{-1} fresh wt.) and *Chnoospora implexa* (0.20±0.02 mg g^{-1} fresh wt.). Compared to red and brown algae, phycoerythrin content was less in green seaweeds. Differences in phycoerythrin content were not significant among the green seaweeds investigated (Table 3).

4.2.8. Crude carbohydrate yield

Crude carbohydrate yield of algae are presented in Table 4. Generally crude carbohydrate yield was more in red and brown seaweeds than green algae. Crude carbohydrate yield was high in *Gracilaria* species(Agarophytes) followed by *Hypnea* species and other red algae and differences between species of the same genus was not very significant. However, significantly, maximum crude carbohydrate yield of 57.7±1.7 % alga dry wt. was recorded in *Gracilaria verrucosa* among the red algae which was the highest in all the three groups of seaweeds investigated. Crude carbohydrate yield of 54.3±1.6 % alga dry wt. recorded in *G. edulis* showed insignificant difference with *G. verrucosa* . Minimum crude carbohydrate yield of 21.1±0.9 % alga dry wt. was observed in *Centroceras clavulatum* among the red seaweeds. High crude carbohydrate yield of 51.1±0.6 % alga dry wt. was recorded in brown *Dictyota dichotoma* which was significantly more than the crude carbohydrate yield of another brown alga *Chnoospora implexa*(33.7±0.4% alga dry wt.). Of the green algae, high crude carbohydrate yield of 29.8 ± 1.9 % alga dry wt. was recorded in *Chaetomorpha linum* which did not show any significant difference between the crude carbohydrate yield of *Ulva lactuca, U.reticulata* and *Caulerpa scalpelliformis* but exhibited significant difference with the rest of the green algae. Low crude carbohydrate yield of 17.4±0.6 % alga dry wt. was recorded in *Cladophora facicularis* which was the lowest among the all seaweeds investigated.

4.2.9 Total carbohydrate

Total carbohydrate content was estimated from the crude carbohydrate extracted in all the seaweeds. Generally, total carbohydrate

content was more in red and brown seaweeds than green algae. Among the red seaweeds, like crude carbohydrate yield, the total carbohydrate content was also high in *Gracilaria* species(Agarophytes) followed by *Hypnea* species and other red algae and differences between the species of the same genus was not quite significant. Maximum total carbohydrate content of 67.4±1.4 % crude carbohydrate of alga was recorded in *Gracilaria edulis* among the red algae which was the highest in all the three groups of seaweeds investigated. Total carbohydrate content of 66.4±3.3 % crude carbohydrate of alga of 65.9±2.4 % crude carbohydrate of alga recorded in *G. canaliculata* and *G.verrucosa,* respectively, showed insignificant difference with *G.edulis.* Minimum total carbohydrate content of 29.6±3.6 % crude carbohydrate of alga was recorded in *Jania rubens* among the red seaweeds. High total carbohydrate content of 47.7 ± 3.4 % crude carbohydrate of alga was recorded in brown *Dictyota dichotoma* but it was it was not significantly more than the total carbohydrate content of another brown alga *Chnoospora implexa* (46.7±2.3 % crude carbohydrate of alga). Among the green seaweeds, high total carbohydrate content of 40.1 ± 2.8 % crude carbohydrate of alga was recorded in *Ulva lactuca* which did not show any significant difference between the total carbohydrate of *U.reticulata* and *Chaetomorpha linum* but they exhibited significant differences between the rest of the green algae. Minimum total carbohydrate content of 28.5 ± 1.9 % crude carbohydrate of alga was recorded in *Cladophora facicularis* was among the all seaweeds investigated which showed insignificant difference with *Enteromorpha flexuosa* and *Caulerpa scalpelliformis* (Table 4).

4.2.10 Total Protein

Total protein content was estimated from the crude carbohydrate extracted in all the seaweeds. Total protein content varied in the individual species of seaweeds among the seaweeds investigated. Of the red seaweeds, significantly high total protein content of 37.7±2.9 % crude carbohydrate of alga was recorded in *Gracilaria verrucosa* among the red algae which was the highest in all the three groups of seaweeds investigated. Differences in total protein content were mostly significant among the red algae. Minimum total protein content of 9.2 ± 2.2 % crude carbohydrate of alga was recorded in *Jania rubens* among the red seaweeds. In two brown seaweeds, significantly higher total protein content was recorded in *Chnoospora implexa* (33.4±2.4 % crude carbohydrate of alga) than *Dictyota dichotoma* (25.3±3.4 % crude carbohydrate of alga). Among the green seaweeds, maximum total protein content of 35.5±3.6 % crude carbohydrate of alga was recorded in *Ulva lactuca* which did not show any significant difference with *Chaetomorpha linum* (34.1±1.2 % crude carbohydrate of alga) but exhibited significant difference between the total protein content of rest of the green algae. Minimum total protein content of 20.2±2.3 % crude carbohydrate of alga was recorded in *Enteromorpha flexuosa* among the green seaweeds investigated (Table 4).

4.2.11 Total amino acids

Total amino acid content was estimated from the crude carbohydrate extracted in all the seaweeds and the total amino acid content varied in the individual species among the seaweeds investigated. Among the red seaweeds, significantly high total amino acid content of 30.3±5.2 % crude carbohydrate of alga was recorded in *Gracilaria verrucosa* among the red algae

which was the highest in all the three groups of seaweeds investigated. Next to *Gracilaria verrucosa,* significantly high total amino acid content of 26.2±3.1 % crude carbohydrate of alga was recorded in *G. edulis* among the red algae. Differences in total amino acid content were mostly significant among the red algae. Minimum total amino acid content of 7.5±4.3 % in crude carbohydrate of alga was recorded in *Jania rubens* among the red seaweeds. In two brown seaweeds, significantly high total amino acid content was recorded in *Dictyota dichotoma* (17.6±5.1 % crude carbohydrate of alga) than *Chnoospora implexa* (14.9±3.2 % crude carbohydrate of alga). Among the green seaweeds, maximum total amino acid content of 26.3±2.1 % crude carbohydrate of alga was recorded in *Ulva lactuca* which did not show significant difference with *Chaetomorpha linum* (25.6±2.1 % crude carbohydrate of alga) and *Ulva reticulata* (24.1±3.7 % crude carbohydrate of alga) but exhibited significant difference with the total amino acid content of rest of the green algae. Minimum total amino acid content of 18.1±4.1 % crude carbohydrate of alga was recorded in *Cladophora facicularis* among the green seaweeds investigated (Table 4).

4.2.12 Total Phenol

Total phenol estimated from the crude carbohydrate extracted from all the seaweeds is presented in the Table 5. Generally, phenolic content was similar and exhibited mostly insignificant differences among the seaweeds in general red seaweeds in particular. Among the red seaweeds, maximum total phenol of 0.51 ± 0.02 % crude carbohydrate of alga recorded in *Gracilaria verrucosa* was significantly higher than the other red algae investigated. Next to *Gracilaria verrucosa,* moderate amount of total phenol of 0.47±0.02 % crude carbohydrate of alga recorded in *G. edulis* did not exhibit difference with *G.*

corticata var. *cylindrica, G. canaliculata, G. foliifera, Hypnea flagelliformis, H.musciformis, H. valentiae , Champia parvula* and *Centroceras clavulatum*. Minimum total phenolic content of 0.17 ± 0.00 % crude carbohydrate of alga was recorded in *Jania rubens* among the red seaweeds. Generally higher phenolic content was recorded in brown algae than red and green algae. In two brown seaweeds, significantly higher total phenol was recorded in *Dictyota dichotoma* (0.68±0.01 % crude carbohydrate of alga) than *Chnoospora implexa* (0.57± 0.01 % crude carbohydrate of alga). Among the green seaweeds, maximum total phenol of 0.41±0.02 % crude carbohydrate of alga was recorded in *Chaetomorpha linum* which did not show any significant difference with *Ulva lactuca*(0.39±0.01 % crude carbohydrate of alga). Minimum total phenolic content of 0.26±0.01 % crude carbohydrate of alga was recorded in *Enteromorpha flexuosa* and did not show any significant difference with *E. intestinalis*.

4.2.13 Water Retention Capacity (WRC)

Water Retention Capacity (WRC) was assessed in crude carbohydrate extracted from all the seaweeds. Generally WRC was higher in seaweeds of commercial value for phycocolloid than other seaweeds. Among the red seaweeds, maximum WRC of 2.89±0.80 g H_2O g^{-1} crude carbohydrate of alga recorded in *Gracilaria canaliculata* was not significantly higher than the WRC of *G. edulis*(2.72±0.51 g H_2O g^{-1} crude carbohydrate of alga) and *G. verrucosa* (2.51±0.66 g H_2O g^{-1} crude carbohydrate of alga). Minimum WRC of 0.73±0.71 g H_2O g^{-1} crude carbohydrate of alga was recorded in *Spiridia hypnoides* among the red seaweeds. In two brown seaweeds, significantly higher WRC was recorded in *Dictyota dichotoma* (1.71±0.55 g H_2O g^{-1} crude carbohydrate of alga)

than *Chnoospora implexa* (1.23±0.42 g H_2O g^{-1} crude carbohydrate of alga).Generally WRC of green algae was less among the seaweeds investigated. Of the green seaweeds, maximum WRC of 1.06±0.73 g H_2O g^{-1} crude carbohydrate of alga was recorded in *Ulva lactuca* which did not show any significant difference with *Ulva reticulata* (0.95±0.71 g H_2O g^{-1} crude carbohydrate of alga) and *Chaetomorpha linum*(0.99±0.75 g H_2O g^{-1} crude carbohydrate of alga). Minimum WRC of 0.78 ± 0.42 g H_2O g^{-1} crude carbohydrate of alga recorded in *Enteromorpha flexuosa* did not show any significant difference with *E. intestinalis, Caulerpa scalpeliformis* and *Cladophora facicularis*(Table 5).

4.2.14 Phycocolloids- Agar, carrageenan and alginate

Agar, carrageenan and alginate were extracted from the crude carbohydrate obtained from some seaweeds of commercial value for phycocolloids. Agar was extracted from the crude carbohydrate of red seaweeds such as *Grateloupia filicina, Gracilaria corticata* var. *corticata, G. corticata* var. *cylindrical, G. edulis , G. canaliculata , G. verrucosa* and *G. foliifera.*Among the agarophytes, significantly higher agar yield of 82.22±5.2 % dry crude carbohydrate of alga was extracted in *G. canaliculata* than the agar yields of *Gracilaria corticata* var. *corticata, G. corticata* var. *cylindrica, G. edulis, G. verrucosa* and *G. foliifera* and differences in agar yield among them insignificant. Carrageenan was extracted from the red algae such as *Grateloupia filicina,Hypnea flgelliformis, H.musciformis* and *H. valentiae.* Among the carrageenophytes, significantly high yield of carrageenan of 74.61 ± 3.3 % dry crude carbohydrate of alga was recorded in *Hypnea musciformis* followed by *H. valentiae, H. flagelliformis* and *Grateloupia filicina.* Alginate yield of 40.42±2.6

% dry crude carbohydrate of alga was recorded only in alginophyte *Dictyota dichotoma* (Table 5).

4.2.15 Sulphate

Sulphate was estimated in the crude carbohydrate extracted from all the seaweeds. Mostly, significant difference in sulphate content was exhibited among the seaweeds. Sulphate content was higher in green seaweeds than red and brown algae. Seaweeds of commercial value for phycocolloids recorded lower sulphate level than other seaweeds. Among the red seaweeds, significantly high sulphate content of 87.32±5.2 mg g^{-1} dry crude carbohydrate of alga was recorded in *Jania rubens.* Moderate level of sulphate content were recorded in agarophytes such as in *Gracilaria canaliculata, G. corticata* var. *corticata, G. corticata* var. *cylindrica, G. edulis, G. verrucosa* and *G. foliifera.* Minimum sulphate content of 18.43 ± 3.8 mg g^{-1} dry crude carbohydrate of alga was recorded in *Hypnea flagelliformis* which showed significant differences than the other red algae investigated. In two brown seaweeds, significantly higher sulphate content was recorded in *Dictyota dichotoma* (55.66±5.8 mg g^{-1} dry crude carbohydrate of alga) than *Chnoospora implexa* (49.25±3.6 mg g^{-1} dry crude carbohydrate of alga). Among the green seaweeds, maximum sulphate content of 109.42±7.9 mg g^{-1} dry crude carbohydrate of alga was recorded in *Chaetomorpha linum* which showed significant difference with *Ulva lactuca*(94.75±9.2 mg g^{-1} dry crude carbohydrate of alga) containing optimum amount of sulphate among the green seaweeds. Minimum sulphate content of 73.83±7.2 mg g^{-1} dry crude carbohydrate of alga was recorded in *Cladophora facicularis* among the green seaweeds (Table 5).

4.3. Seasonal variation in proximate composition of seaweeds

Results on the biochemical characteristics such as dry wt., total chlorophyll and total carbohydrate content of two red algae *Gracilaria edulis* and *G. verrucosa* and two green algae *Ulva lactuca* and *Chaetomorpha linum* recorded during four different seasons from four stations in the year 2007 are presented in the Table 6,7 and 8.

4.3.1 Dry wt.

Seasonal variations in the dry wt. content of seaweeds are presented in the Table 6. Generally, dry wt. content was more in red seaweeds than green seaweeds and algae collected from Thondi and Theerthandathanam showed higher dry wt. than Nambutalai and MR Pattanam. Among the seasons, maximum dry wt. was recorded from algae collected during monsoon season followed by post-monsoon, pre-monsoon and summer seasons and differences in dry wt. of seaweeds collected during the four seasons from each station were mostly significant. Maximum significant dry wt. of 157.4 ± 11.5 mg g^{-1} fresh wt. was recorded in red alga *Gracilaria verrucosa* collected at Theerthandathanam (Station-IV) during monsoon season among the seaweeds collected from four stations whereas significantly low dry wt. of 102.1 ± 9.1 mg g^{-1} fresh wt. was recorded at MR Pattanam (Station-III) collection during summer season. In *Gracilaria edulis,* significantly high dry wt. of 156.9 ± 7.7 mg g^{-1} fresh wt. was recorded at Theerthandathanam(Station-IV) during monsoon season whereas significantly low dry wt. of 102.1 ± 9.1 mg g^{-1} fresh wt. was recorded at MR Pattanam (Station-III) collection during summer season. Significantly high dry wt. of 97.5 ± 10.5 mg g^{-1} fresh wt. and 109.2 ± 9.7 mg g^{-1} fresh wt. were

recorded at Theerthandathanam (Station-IV) during post-monsoon season whereas low dry wt. of 71.5 ± 10.1 mg g^{-1} fresh wt. and 69.2 ± 10.8 mg g^{-1} fresh wt. were recorded at MR Pattanam (Station-III) during summer season in *Ulva lactuca* and *Chaetomorpha linum*, respectively.

4.3.2 Total chlorophyll

Seasonal variations in the total chlorophyll of seaweeds are presented in the Table 7. Unlike dry wt. of algae, total chlorophyll content was more in green seaweeds than red seaweeds and algae collected from Nambutalai and MR pattanam showed higher total chlorophyll content than Thondi and Theerthandathanam. Among the seasons, maximum significant total chlorophyll was recorded mostly from algae collected during monsoon season followed by post-monsoon, pre-monsoon and summer seasons and the differences in dry wt. of seaweeds collected during the four seasons from each station were mostly significant. Maximum significant total chlorophyll of 0.79 ± 0.05 mg g^{-1} fresh wt. was recorded in green alga *Ulva lactuca* collected at Nambutalai (Station-I) during monsoon season among the seaweeds collected from the four stations whereas significantly low dry wt. of 0.43 ± 0.07 mg g^{-1} fresh wt. was recorded at Theerthandathanam(Station-IV) collection during summer season. In *Chaetomorpha linum,* significantly high dry wt. of 0.75 ± 0.04 mg g^{-1} fresh wt. was recorded at Nambutalai(Station-I) during monsoon season whereas significantly low dry wt. of 0.41 ± 0.04 mg g^{-1} fresh wt. was recorded at Theerthandathanam(Station-IV) collection during summer season. In *Gracilaria edulis*, significantly high total chlorophyll of 0.58 ± 0.05 mg g^{-1} fresh wt. was recorded at MR Pattanam (Station-III) during monsoon season whereas significantly low total chlorophyll of 0.32 ± 0.03 mg g^{-1} fresh wt. was recorded

at Thondi (Station-II) collection during summer season. High total chlorophyll of 0.59 ± 0.06 mg g $^{-1}$ fresh wt. was recorded at Thondi (Station-III) during monsoon season which showed insignificant difference during post-monsoon season and low total chlorophyll of 0.32 ± 0.04 mg g $^{-1}$ fresh wt. was recorded at Theerthandathanam(Station-IV) collection during the summer season but was not significant with the pre-monsoon collection in red alga *Gracilaria verrucosa*.

4.3.3 Total carbohydrate

Seasonal variations in the total carbohydrate content of seaweeds are presented in the Table 8. Like dry wt., generally, total carbohydrate was more in red seaweeds than green seaweeds and algae collected from Thondi and Theerthandathanam showed higher total carbohydrate than Nambutalai and MR pattanam. Among the seasons, maximum total carbohydrate was recorded from algae collected during monsoon season followed by post-monsoon, pre-monsoon and summer seasons and the differences in total carbohydrate of seaweeds collected during the four seasons from each station were mostly significant. Maximum significant total carbohydrate of 55.7 ± 6.2 mg g $^{-1}$ fresh wt. was recorded in red alga *Gracilaria edulis* collected at Theerthandathanam (Station-IV) during the monsoon season among the seaweeds collected from four stations whereas significantly low total carbohydrate of 22.4 ± 8.9 mg g $^{-1}$ fresh wt. was recorded at Nambutalai (Station-I) collection during the summer season. In *Gracilaria verrucosa*, high total carbohydrate of 55.3 ± 8.5 mg g $^{-1}$ fresh wt. was recorded at Theerthandathanam (Station-IV) during monsoon season whereas significantly low total carbohydrate of 20.1 ± 8.3 mg g $^{-1}$ fresh wt. was recorded at MR Pattanam (Station-III) collection during the summer

season. High total carbohydrate of 52.1 ± 9.8 mg g $^{-1}$ fresh wt. and 53.8 ± 8.4 mg g $^{-1}$ fresh wt. was recorded at Theerthandathanam (Station-IV) during the monsoon season and low total carbohydrate of 33.5 ± 5.2 mg g $^{-1}$ fresh wt. and 37.3± 6.3 mg g $^{-1}$ fresh wt. was recorded at MR Pattanam (Station-III) during summer season in *Ulva lactuca* and *Chaetomorpha linum*, respectively (Table 8).

4.4.1 Monthly biochemical characteristics of selected seaweeds and Thondi meteorological parameters

Monthly meteorological parameters such as mean maximum temperature, mean minimum temperature, mean relative humidity, mean total rainfall and mean wind speed of Thondi coast recorded every month from October 2006 to September 2007 for a year(Table 9) were compared with dry wt., total chlorophyll and total carbohydrate recorded on full moon day of every month from red alga *Gracilaria verrucosa* and green alga *Chaetomorpha linum* found along the Thondi coast where the meteorological data made from October 2006 to September 2007 for a year(Table 10). Generally mean maximum temperature was high during the months of summer season followed by the months of pre-monsoon, post-monsoon and monsoon. Highest mean maximum temperature of 34.1^{0}C was recorded during the month of September 2007(Pre-monsoon) and lowest mean maximum temperature of 30.0^{0}C was recorded during the month of November2006 (Monsoon). As like mean maximum temperature, mean minimum temperature was high during months of summer season followed by the months of pre-monsoon, post-monsoon and monsoon. Highest mean minimum temperature of 28.0 ^{0}C was recorded during the month of May

2007(summer) and lowest mean minimum temperature of 21.9 ^{0}C was recorded during the month of January 2007(post-monsoon). High mean relative humidity was recorded during months of monsoon season followed by months of post-monsoon, pre-monsoon and summer. Highest mean relative humidity of 87.0 was recorded during the month of November 2006(monsoon) and lowest mean relative humidity of 73.0 was recorded during the month of September 2007(Pre-monsoon). High mean total rainfall was recorded during the months of monsoon season followed by months of post-monsoon, pre-monsoon and summer. Maximum mean total rainfall of 367.7 mm was recorded during the month of October 2006(monsoon) and minimum mean total rainfall of 2.2 mm was recorded during March 2007(Post-monsoon). In the month of May 2007(summer), no rainfall was recorded. High mean wind speed was recorded during the months of summer followed by the months of pre-monsoon, post-monsoon and monsoon. Maximum mean wind speed of 13.0 kmph was recorded during the month of May 2007(summer) and minimum mean wind speed of 4.0 kmph was recorded during November 2006(monsoon).

Generally dry wt. and total carbohydrate were more in red alga *Gracilaria verrucosa* than green alga *Chaetomorpha linum* whereas total chlorophyll was more in latter than former. Dry wt. and total carbohydrate were higher in the months of monsoon and post-monsoon than summer and pre-monsoon in both seaweeds whereas total chlorophyll recorded similar trend in all months in a year. Mean relative humidity and mean total rain fall were higher during the months of monsoon and post-monsoon seasons where these seasons' dry wt. and total carbohydrate of algae were high. Dry wt. and

total carbohydrate of algae recorded low during the months of summer and pre-monsoon where mean relative humidity and mean total rain fall were also less and mean maximum temperature, mean minimum temperature, and mean wind speed were high. Similar trend in total chlorophyll in algae in all the months in a year showed that increase in the mean relative humidity and mean total rain fall and decrease in the mean maximum temperature, mean minimum temperature, and mean wind speed during the months of monsoon and post-monsoon; these meteorological parameters were vice versa during the months of summer and pre-monsoon did not limit the total chlorophyll in the algae(Table 10).

4.4.2 Correlation matrix of monthly biochemical characteristics of seaweeds to meteorological parameters

Correlation between monthly meteorological parameters and dry wt., total chlorophyll and total carbohydrate of red alga *Gracilaria verrucosa* and green alga *Chaetomorpha linum* was arrived. Mean maximum temperature showed significant positive correlation with mean wind speed and total carbohydrate whereas with mean minimum temperature, mean relative humidity and mean total rainfall correlation was negative but with dry wt. and total chlorophyll showed insignificant positive correlation. Mean minimum temperature showed significant positive correlation with mean relative humidity, dry wt. total chlorophyll and total carbohydrate and insignificant positive correlation with wind speed and total rainfall. Mean relative humidity minimum temperature showed significant positive correlation with total rain fall, dry wt. total chlorophyll and total carbohydrate and significant negative correlation with wind speed. Total rain fall showed significant positive

correlation with dry wt. total chlorophyll and total carbohydrate and insignificant negative correlation with wind speed. Mean wind speed showed insignificant negative correlation with dry wt. total chlorophyll and total carbohydrate. Highly significant positive correlation was observed between dry wt., total chlorophyll and total carbohydrates in seaweeds (Table 11).

4.5 Amino acids profile of red seaweeds *Gracilaria edulis* and *G.verrucosa*; and green seaweeds *Ulva lactuca* and *Chaetomorpha linum* obtained through HPLC

Results on amino acids profiles of two red seaweeds *Gracilaria edulis*(Fig. 3) and *G.verrucosa*(Fig. 4); and two green seaweeds *Ulva lactuca*(Fig.5) and *Chaetomorpha linum*(Fig. 6) obtained through HPLC are presented in the Table 12.

In the present investigation, two red and two green seaweeds were evaluated for various amino acids using HPLC. Depending on the species, quantity of each amino acid varied. Of the total amino acids, non-essential amino acids were higher than essential amino acids in all 4 seaweeds investigated. However, number of essential amino acids was 9 whereas non-essential amino acids were 8. Essential amino acids recorded were Tryptophan, Threonine, Valine, Lysine, Isoleucine, Leucine, Phenylalanine, Arginine, Histidine and Methionine and non-essential amino acids were Aspartic acid, Serine, Glutamic acid , Glycine, Alanine ,Tyrosine, Asparagines and Cysteine from four seaweeds. Among the 4 seaweeds, a maximum of total and essential amino acids were recorded in *Gracilaria verrucosa* followed by *Ulva lactuca, Gracilaria edulis* and *Chaetomorpha linum* respectively. Of the 9 essential amino acids, leucine recorded highest of 2.22 g 100 g $^{-1}$dry wt. in *Gracilaria verrucosa* followed by *Ulva lactuca* (1.83

g 100 g $^{-1}$dry wt.), *Gracilaria edulis*(1.52 g 100 g $^{-1}$dry wt.) and *Chaetomorpha linum*(1.03 g 100 g $^{-1}$dry wt.) respectively. Like leucine, a maximum of Valine (1.99 g 100 g $^{-1}$dry wt.), Phenylalanine(1.96 g 100 g $^{-1}$dry wt.), Threonine(1.79 g 100 g $^{-1}$dry wt.), Isoleucine (1.02 g 100 g $^{-1}$dry wt.), Histidine(0.99 g 100 g $^{-1}$dry wt.) and Methionine(0.89 g 100 g $^{-1}$dry wt.) were observed in red seaweed *Gracilaria verrucosa* in decreasing order whereas maximum Arginine (0.84 g 100 g $^{-1}$dry wt.) was found in *G. edulis.* Maximum Lysine(1.21 g 100 g $^{-1}$dry wt.) was recorded in green alga *Ulva lactuca*. Among 4 seaweeds, a maximum of non-essential amino acids were recorded in *Gracilaria verrucosa* followed by *Ulva lactuca*, *Chaetomorpha linum* and *Gracilaria edulis* respectively. Among 8 non-essential amino acids, Glutamic acid recorded highest of 3.10 g 100 g $^{-1}$dry wt. in *Gracilaria verrucosa* followed by *Ulva lactuca*(2.94 g 100 g $^{-1}$dry wt.), *Chaetomorpha linum*(2.77 g 100 g $^{-1}$dry wt.) and *Gracilaria edulis*(2.33 g 100 g $^{-1}$dry wt.) respectively. Like Glutamic acid in *Gracilaria verrucosa*, high amount of other non-essential amino acids recorded was Aspartic acid (2.77 g 100 g $^{-1}$dry wt.). Maximum of Alanine(1.93 g 100 g $^{-1}$dry wt.), Cysteine (1.31 g 100 g $^{-1}$dry wt.) and Serine (1.18 g 100 g $^{-1}$dry wt.) were observed in green seaweed *Ulva lactuca* whereas in another green alga *Chaetomorpha linum*, Glycine (2.01 g 100 g $^{-1}$dry wt.), Tyrosine(1.22 g 100 g $^{-1}$dry wt.) and Asparagine(1.11 g 100 g $^{-1}$dry wt.) recorded maximum level(Table 12).

4.6 Evaluation of Immunostimulation potential of seaweeds under *in vivo*

Results on red blood cells(RBC), White blood cells(WBC), neutrophils, neutrophil adhesive and total protein of blood samples of immunosuppressive rats fed with powdered samples and crude amino acids

extracts of *Gracilaria edulis, G. verrucosa, Ulva lactuca* and *Chaetomorpha linum* are presented in the Table 13.

4.6.1 RBC

Blood samples of immunosuppressive rats fed with powdered samples and crude amino acids extracts of seaweeds resulted significant increase in RBC count compared to rats of immunosuppressive control. However, increase in RBC in the blood samples of immunosuppressive rats fed with powdered samples of seaweeds was not significant compared to control rats (administered only with saline) but exhibited significant increase in the RBC of blood samples of immunosuppressive rats fed with crude amino acids extracts of seaweeds. Maximum increase in RBC of 5.77 ± 0.56 million^{-1}µl was recorded in the blood samples of immunosuppressive rats fed with crude amino acid extracts of *Ulva lactuca* but differences of RBC in the blood samples of immunosuppressive rats fed with crude amino acid extracts of seaweeds was not significant but showed significant difference compared to blood samples of immunosuppressive rats fed with powdered seaweed samples. However, high RBC of 3.99 ± 0.57 million^{-1}µl recorded in the blood samples of immunosuppressive rats fed with powered samples of *Ulva lactuca* was not significant compared to the RBC of blood samples of immunosuppressive rats fed with other powdered samples of seaweeds and control rats (administered only with saline) but they exhibited significant difference compared to rats of immunosuppressive control which recorded minimum RBC of 1.05 ± 0.11 million^{-1}µl in the blood samples (Table 13).

4.6.2 WBC

Like RBC, WBC in the blood samples of immunosuppressive rats fed with powdered samples and crude amino acids extracts of seaweeds resulted significant increase compared to rats of immunosuppressive control. However, increasing in WBC in the blood samples of immunosuppressive rats fed with powdered samples of seaweeds did not significant compared to control rats (administered only with saline) but exhibited significant increase in the WBC of blood samples of immunosuppressive rats fed with crude amino acids extracts of seaweeds. Maximum increase in WBC of 8.67 ± 0.37 thousand^{-1} µl recorded in the blood samples of immunosuppressive rats fed with crude amino acid extracts of *Ulva lactuca* was significant compared to WBC in the blood samples of immunosuppressive rats fed with other crude amino acid extracts of seaweeds but showed significant difference compared to blood samples of immunosuppressive rats fed with powdered seaweed samples. However, high WBC of 5.48 ± 0.91 thousand^{-1} µl recorded in the blood samples of immunosuppressive rats fed with powdered samples of *Ulva lactuca* was not significant compared to the WBC of blood samples of immunosuppressive rats fed with other powdered samples of seaweeds and control rats (administered only with saline) but they exhibited significant difference compared to rats of immunosuppressive control which recorded minimum WBC of 3.28 ± 0.34 thousand^{-1} µl in the blood samples (Table 13).

4.6.3 Neutrophils

Neutrophils in the blood samples of immunosuppressive rats fed with powdered seaweed samples and crude amino acids extracts of seaweeds showed significant increment equal to control rats (administered

only with saline) compared to rats of immunosuppressive control as like RBC and WBC. However, neutrophils in the blood samples of immunosuppressive rats fed with powdered seaweed samples were more than in immunosuppressive rats fed with crude amino acids extracts of seaweeds . Maximum neutrophils of 23.57 ± 0.27 thousand^{-1} µl recorded in the blood samples of immunosuppressive rats fed with powdered sample of *Ulva lactuca* was not significant compared to neutrophils in the blood samples of immunosuppressive rats fed with other powdered samples of seaweeds and control rats (administered only saline) but showed significant differences compared to blood samples of immunosuppressive rats fed with crude amino acids of seaweeds. However, increase in neutrophils in the blood samples of immunosuppressive rats fed with crude amino acids of seaweeds was significant compared to rats of immunosuppressive control whereas increase in neutrophils was significantly low compared to control rats (administered only with saline). Among the immunosuppressive rats fed with crude amino acids extracts of seaweeds, high neutrophils of 15.39 ± 1.45 thousand^{-1} µl recorded in the blood samples of immunosuppressive rats fed with crude amino acid extracts of *Ulva lactuca* did not significant compared with neutrophils in the blood samples of immunosuppressive rats fed with other crude amino acid extracts of seaweeds. Minimum neutrophils of 12.40 ± 0.51thousand^{-1} µl was recorded in the blood samples of rats of immunosuppressive control (Table 13).

4.6.4 Neutrophil adhesive

Results on the neutrophil adhesive in the blood samples of rats are presented in the Table 13. Compared to normal rats, immunosuppressive

rats and immunosuppressive rats fed with powdered samples and crude amino acids extracts of seaweeds showed significant decrease in neutrophil adhesive in the blood samples. Even though immunosuppressive rats fed with powdered samples and crude amino acids extracts of seaweeds showed increase in neutrophil adhesive in the blood samples, the increment did not show significant compared to immunosuppressive control rats and immunosuppressive rats fed with crude amino acid extracts of red seaweeds *Gracilaria edulis* and *G. verrucosa*. Maximum neutrophil adhesive of 33.56 ± 0.23 % was recorded in the blood samples of normal control rats(administered only with saline) and showed insignificant difference in the neutrophil adhesive in the blood sample of immunosuppressive rats fed with powdered sample of red *Gracilaria verrucosa* (30.18 ± 0.21 %) and green *Ulva lactuca*(31.31 ±0.62 %).

4.6.5 Total protein

Total protein in the blood samples of immunosuppressive rats fed with powdered seaweed samples and crude amino acids extracts of seaweeds showed significant increment compared to rats of immunosuppressive control and showed insignificant difference compared to control rats (administered only with saline).Differences of total protein content in the blood sample of immunosuppressive rats fed with powdered samples and crude amino acids extracts of seaweeds were not significant. Maximum total protein content of 6.01 ± 0.52 mg/dl was recorded in the blood samples of immunosuppressive rats fed with crude amino acid extracts of *Ulva lactuca.* Minimum total protein content of 3.02 ± 0.61 mg/dl was recorded in the blood samples of immunosuppressive control rats(Table 13).

4.6.6 Ig G

IgG level in the blood serum of rats fed with powdered samples and crude amino acids extracts of seaweeds appeared on the gel were detected based on intensity of band. Blood serum of control rats (administered only with saline) exhibited strong Ig G protein band whereas immunosuppressive rats and immunosuppressive rats fed with crude amino acids extracts of green algae *Ulva lactuca* and *Chaetomorpha linum*; and powdered sample of red *Gracilaria edulis* did not develop Ig G protein. Blood sample of rats fed with powdered sample of *Ulva lactuca* exhibited high IgG protein as strong band. Blood sample of rats fed with amino acid extracts of *Gracilaria edulis* exhibited optimum level of IgG protein whereas amino acid extracts of *Gracilaria verrucosa* and powder of *Gracilaria verrucosa* and *Chaetomorpha linum* exhibited moderate level of IgG protein(Plate VII).