3. MATERIALS AND METHODS

A field experiment entitled **"Effect of Phosphorus, Sulphur and Seaweed Sap on Productivity of Chickpea (*Cicer arietinum* L.)"** was carried out during two consecutive *rabi* seasons of 2012-13 and 2013-14. The details of experimental techniques adopted, criteria used for treatment evaluation and methods followed during the course of investigation are described in this chapter.

3.1 EXPERIMENTAL SITE

The experiment was conducted at the Instructional Farm, College of Technology and Engineering, Udaipur during *rabi* seasons of 2012-13 and 2013-14. The site is situated in South-Eastern part of Rajasthan at the altitude of 582.17 metre above mean sea level with 24º35' N latitude and 73°42' E longitude. This region falls under agro-climatic zone IVa "Sub-humid Southern Plain and Aravalli Hills" of Rajasthan. The experiment was conducted in the same field during both the years of study.

3.1.1 Climate and weather condition

This zone has typical sub-tropical climatic conditions characterized by mild winters and moderate summer associated with high relative humidity during the months of July to September. The mean annual rainfall of the region is 637 mm, most of which is contributed by south-west monsoon from July to September.

The mean weekly meteorological parameters recorded at meteorological observatory, College of Technology and Engineering, Udaipur during crop periods are presented in Table 3.1 and depicted in Fig. 3.1a and 3.1b. These observations reveal that maximum and minimum temperatures ranged between 22.1 to 33.1 and 1.3 to 14.0 during 2012-13 and 23.2 to 32.8 and 3.7 to 16.2 ºC during 2013-14, respectively. The maximum and minimum relative humidity ranged between 78 to 94 and 24 to 52 during 2012-13 and 84 to 99 and 23 to 82 per cent during 2013-14, respectively. The total rainfall received during the chickpea crop season of the 2012-13 was 1.4 mm and while 17.1 mm rains were received during 2013-14. The evaporation from the USWB class-A pan evaporimeter during the corresponding crop season ranged from 1.6 to 3.8 and 1.0 to 3.4 mm day^{-1}, and total evaporation during crop season were, 47.0 and 40.1 mm, respectively.

Table 3.1. Mean weekly meteorological data during crop growing season (2012-13 and 2013-14)

Standard Meteorological Week No. (SMW)	Date	Temperature (°C)				Relative humidity (%)				Total rainfall (mm)		Wind velocity (km h^{-1})		Evaporation (mm day^{-1})		Sunshine (hrs)	
		Maximum		Minimum		Maximum		Minimum									
		2012-13	2013-14	2012-13	2013-14	2012-13	2013-14	2012-13	2013-14	2012-13	2013-14	2012-13	2013-14	2012-13	2013-14	2012-13	2013-14
42	15 Oct.-21 Oct.	33.1	32.8	14.0	16.2	94	94	29	63	0	0	0.8	1.3	3.8	3.1	8.7	6.0
43	22 Oct.-28 Oct.	31.3	31.3	12.1	12.5	82	91	34	60	0	0	0.9	0.9	3.4	3.4	9.1	9.0
44	29 Oct.-4 Nov.	28.9	32.0	8.5	12.6	92	91	35	50	0	0	1.7	1.3	2.0	2.6	7.6	7.5
45	5 Nov.-11 Nov.	29.0	28.1	8.1	13.8	94	87	38	57	0	0	0.8	1.0	2.4	2.6	8.0	6.8
46	12 Nov.-18 Nov.	29.1	26.3	9.4	8.7	89	95	38	69	0	0	0.8	1.0	2.6	2.0	8.1	7.4
47	19 Nov.-25 Nov.	27.8	27.8	8.7	6.5	88	90	32	56	0	0	0.8	0.7	2.0	2.1	8.3	8.3
48	26 Nov.-2 Dec.	27.4	30.7	7.5	7.9	83	93	32	59	0	0	0.9	0.9	2.2	2.0	7.8	8.2
49	3 Dec.-9 Dec.	29.2	26.7	8.7	6.5	91	95	36	72	0	0	0.7	0.9	2.0	2.1	8.0	6.6
50	10 Dec.-16 Dec.	27.0	27.8	8.5	5.7	89	95	26	73	0	0	1.2	0.6	2.0	1.6	7.8	8.7
51	17 Dec.-23 Dec.	25.1	26.1	5.4	4.0	87	96	44	76	0	0	1.2	0.8	1.9	1.3	7.8	8.1
52	24-31 Dec.	24.9	23.2	4.2	5.2	90	96	37	82	0	0	0.9	0.8	1.7	1.2	7.4	5.6
1	1 Jan.-7 Jan.	22.4	23.6	2.6	5.5	89	98	29	77	0	0	0.8	1.1	1.6	1.0	7.7	6.3
2	8 Jan.-14 Jan.	25.4	23.2	3.8	3.7	87	97	33	71	0	0	1.0	1.6	1.9	1.0	8.3	5.9
3	15 Jan.-21 Jan.	24.2	23.6	4.6	4.7	88	99	34	59	0	16.6	2.0	1.3	2.0	1.0	7.8	7.8
4	22 Jan.-28 Jan.	22.1	24.2	1.3	5.6	87	97	33	45	0	0.5	1.6	1.0	2.5	1.1	9.1	5.3
5	29 Jan.-4 Feb.	25.8	26.9	7.8	6.0	87	96	37	39	0	0	0.7	0.8	1.9	1.9	5.9	9.0
6	5 Feb.-11 Feb.	24.1	28.0	5.9	6.7	84	94	52	33	1.2	0	2.4	2.3	2.1	2.3	8.9	8.8
7	12 Feb.-18 Feb.	26.2	24.3	10.3	5.3	85	89	44	42	0.2	0	2.7	2.1	2.4	2.2	8.9	6.7
8	19 Feb.-25 Feb.	26.5	28.0	9.3	6.5	81	91	38	31	0	0	2.5	2.2	3.0	2.6	8.6	6.7
9	26 Feb.-4 March	28.7	27.2	7.0	7.4	78	84	24	23	0	0	2.5	1.7	3.6	3.0	10.0	8.3

Source: Meteorological Observatory, College of Technology and Engineering, Udaipur

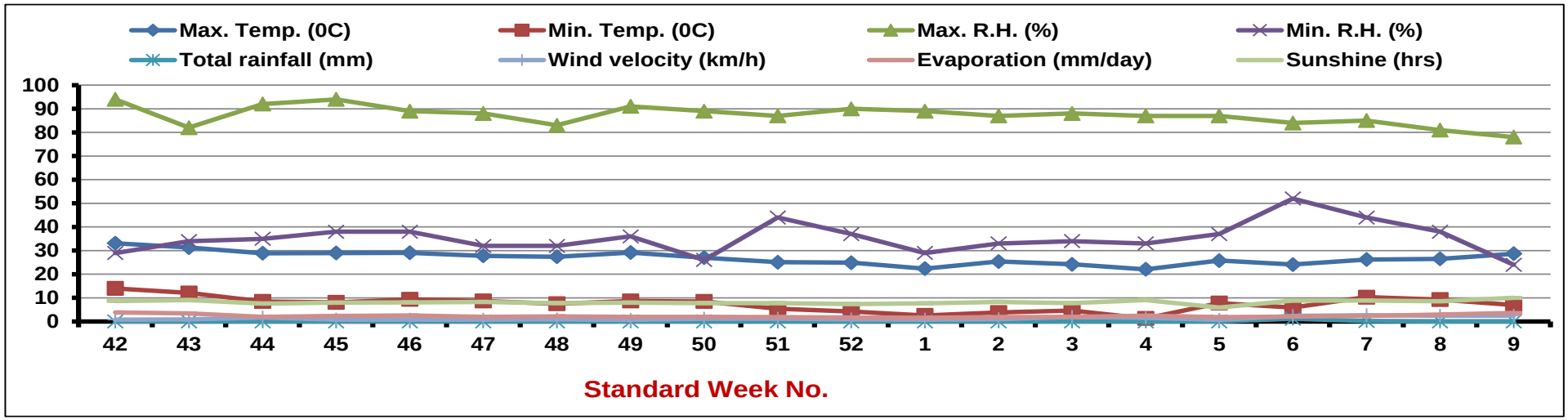


Fig. 3.1(a) Mean weekly meteorological data during crop growing season 2012-13

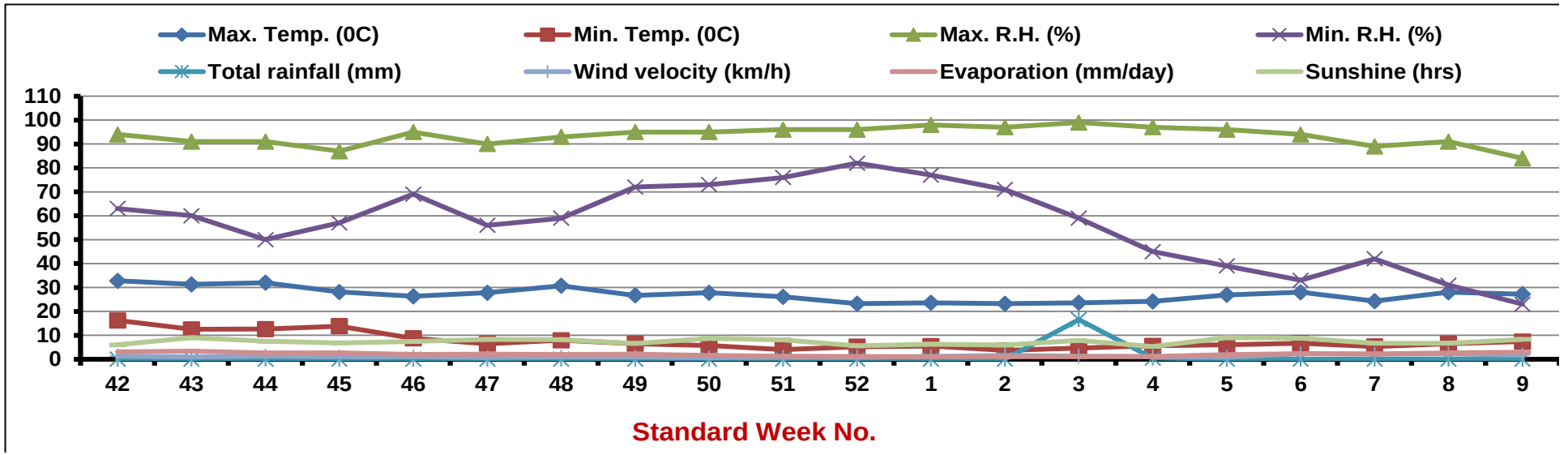


Fig. 3.1(b) Mean weekly meteorological data during crop growing season 2013-14

3.1.2 Physico-chemical properties of experimental soil

In order to ascertain the physico-chemical characteristics of the soil, surface soil (0-15 cm depth) samples were randomly drawn and collected from different spots of the experimental field in both the years. Representative composite samples obtained from the samples of each year, were subjected to physical and chemical analysis separately. The physico-chemical characteristics of the soil of experimental field along with the methods followed for analysis are given in Table 3.2. From the data, it is evident that soil of the site was sandy clay loam in texture, alkaline in reaction and medium in organic carbon status. Further, the soil was low in available nitrogen, phosphorus, sulphur and high in available potassium during the year 2012-13 and 2013-14.

Table 3.2 Physico-chemical characteristics of the experimental soil

Soil properties	**Content**		**Methods used with references**
	2012	**2013**	
A. Mechanical			
Sand (%)	50.73	50.53	International pipette method (Piper, 1950)
Silt (%)	21.43	21.24	
Clay (%)	27.84	28.23	
Textural class	Sandy clay loam	Sandy clay loam	Triangular diagram (Brady and Weil, 2008)
B. Physical			
Bulk density (Mg m^{-3})	1.56	1.52	Core sampler method (Piper, 1950)
Particle density(Mg m^{-3})	2.64	2.66	Method No.33, USDA Hand book No. 60 (Richard,1954)
Total porosity (%)	40.91	42.86	USDA Hand book-60 (Richard, 1954)
C. Chemical			
Organic carbon (g kg^{-1})	6.80	6.70	Walkley and Black's wet digestion method (Walkley and Black, 1947)
EC (dS m^{-1}at 25^0C) (1:2 soil:water suspension)	0.88	0.89	Method No. 4, USDA Hand Book No.-60 (Richard, 1954)
pH (1:2 soil:water suspension)	7.96	7.98	Method No. 21 (b), USDA Hand Book No. 60 (Richard, 1954)
CEC [cmol(p^+)kg^{-1}]	13.60	13.65	Neutral normal ammonium acetate method (Metson, 1956)
Available N (kg ha^{-1})	249.32	238.72	Alkaline permanganate method (Subbiah and Asija, 1956)
Available P (kg ha^{-1})	14.03	14.56	Olsen's method (Olsen *et al.,* 1954)
Available K (kg ha^{-1})	338.45	342.15	Flame photometric method (Richard, 1954)
Available S (mg kg^{-1})	8.9	8.8	Calcium chloride extractable method (Williams and Steinberg, 1959)

3.2 CROPPING HISTORY

The cropping history of the experimental field for the last three years is given in Table 3.3. The experimental field was under continuous cropping for the last four years. In general groundnut in *kharif* and chickpea in *rabi* season was grown for the last four years but in 2011-12 maize-wheat was followed.

Table 3.3 Cropping history of the experimental field

Years	Season	
	kharif	***rabi***
2010-11	Groundnut	Chickpea
2011-12	Maize	Wheat
2012-13	Groundnut	Chickpea*
2013-14	Groundnut	Chickpea*

* Experimental crop

3.3 EXPERIMENTAL DETAILS

3.3.1 Treatments

I. Main plot

A. Phosphorus levels (P_2O_5 kg ha^{-1})

(i) 20 P_1

(ii) 40 P_2

(iii) 60 P_3

B. Sulphur levels (S kg ha^{-1})

(i) 00 S_0

(ii) 20 S_1

(iii) 40 S_2

II. Sub plot

Seaweed sap sprays

(i) Control (Water spray) F_0

(ii) *Kappaphycus* sap 10% F_1

(iii) *Gracilaria* sap 10% F_2

3.3.2 Other experimental details

(i)	Year	: 2012-13 and 2013-14
(ii)	Season	: *rabi*
(iii)	Total Number of treatments combination	: (3 x 3) x 3 = 27
(iv)	Number of replications	: 3
(v)	Total number of plots	: 81
(vi)	Experimental Design	: Split plot design (Phosphorus and sulphur levels in main plots and foliar sprays in sub plots)
(vii)	Plot size –	
	i. Gross	: 5.0 m x 3.6 m = 18.0 m^2
	ii. Net	: 4.0 m x 3.0 m = 12.0 m^2
(viii)	Test crop	: Chickpea
(ix)	Variety	: Pratap channa-1
(x)	Spacing	: 30 cm x 10 cm
(xi)	Seed rate	: 80 kg ha^{-1}

3.3.3 Sources and application of nutrients

The sources used for supplying N and P were urea and DAP, respectively. Mineral gypsum was used to supply S. Complete dose of N, P and S were applied before the sowing as basal application in furrows.

3.4 CHARACTERISTICS OF VARIETY

The variety Pratap Channa-1 (ICCV-88202) is bold seeded, early to medium in maturity having yield potential of 12-14 q ha^{-1}. It is suitable for rainfed areas of Southern Rajasthan.

3.5 DETAILS OF CROP RAISING

Details of field operations carried out for chickpea are given in Table 3.4.

Table 3.4 Schedule of operations during *rabi* 2012-13 and 2013-14

S. No.	Particulars	Date of operations		Remarks
		2012-13	2013-14	
1	Ploughing and planking	18.10.12	28.10.13	Tractor drawn disc harrow and planker
2	Layout of experimental field	21.10.12	31.10.13	Manually
3	Fertilizers application	22.10.12	01.11.13	Basal application manually
4	Sowing of seeds	22.10.12	01.11.13	Manually
5	Seaweed sap spray 1st 2nd 3rd	 21.11.12 06.12.12 21.12.12	 30.11.13 15.12.13 30.12.13	By knapsack sprayer
6	Thinning, hoeing and weeding (i) Thinning (ii) Weeding	 06.11.12 26.12.12	 16.11.13 05.01.14	 Manually Manually
7	Irrigation 1st irrigation 2nd irrigation	 22.10.12 20.01.13	 01.11.13 01.01.14	 By check basin method
8	Harvesting	15.02.13	26.02.14	Manually
9	Threshing and Winnowing	25.02.13	08.03.14	Manually

3.5.1 Field preparation

The experimental field was ploughed thoroughly by tractor drawn disc plough followed by cross harrowing and planking. Thereafter, the field was laid out manually into plots according to the plan of layout (Fig 3.2).

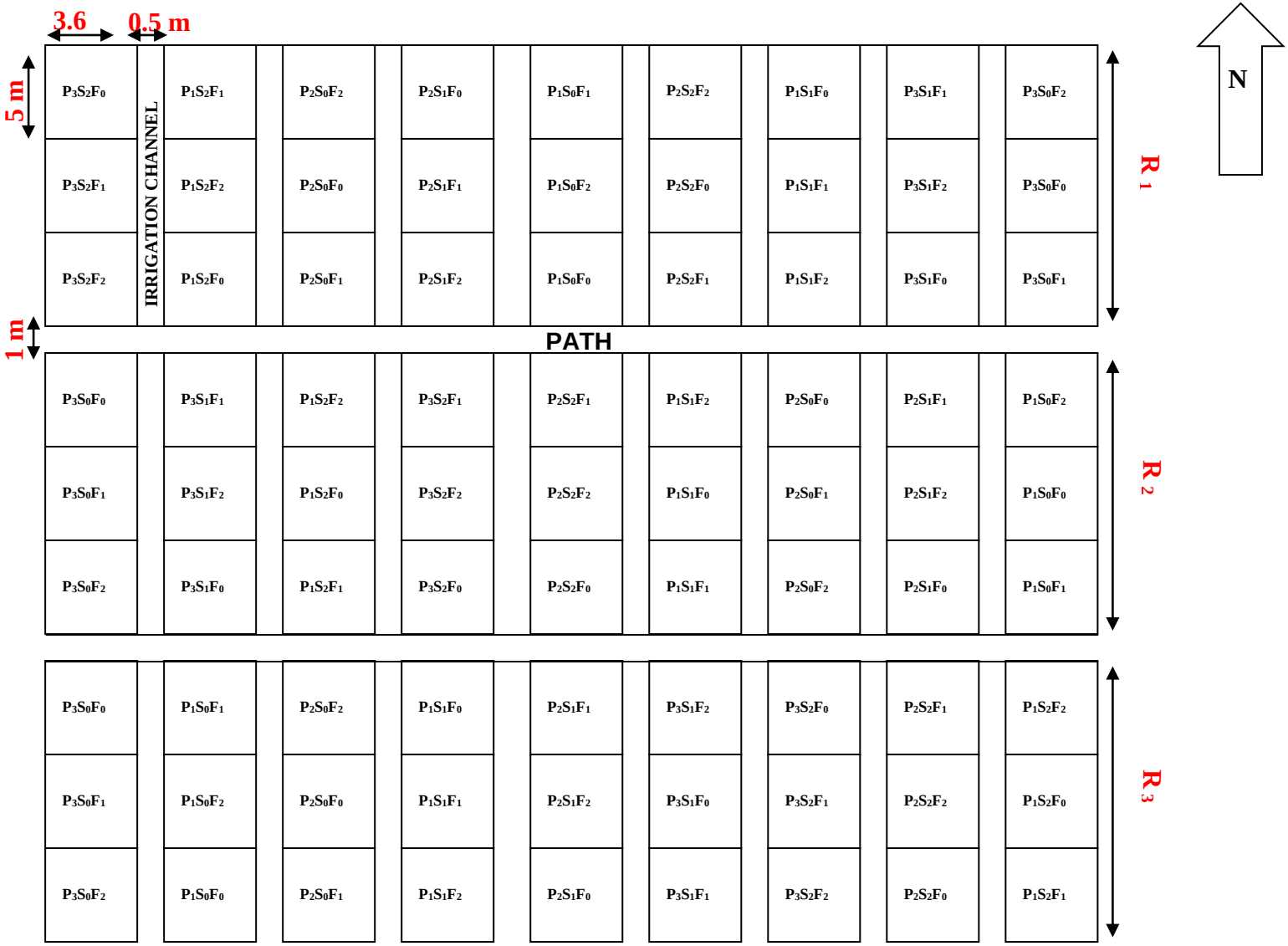
3.5.2 Treatment application

The recommended dose of nitrogen (20 kg ha^{-1}) and phosphorus and sulphur were applied as per treatment and plan of layout. This basal dose of fertilizers *viz*., urea, DAP and gypsum were applied as per requirement of different phosphorus and sulphur treatments in furrows of 10-12 cm depth and at 30 cm distance. After treatment application, the furrows were covered with soil to 3-4 cm and later the same were used for sowing.

Foliar spray of seaweed saps were used as aqueous sprays volume of 600 litre ha^{-1} with the help of knapsack sprayer using solid cone nozzle. The spray of seaweed saps *viz*., *Kappaphycus alvarezii* and *Gracilaria edulis* saps were applied as per treatment concentrations. The chemical composition of seaweed saps as reported by Pramanick *et al.* (2013) are depicted in Table 3.5. In order to make the spray retention effective Teepol, a sticking agent was mixed at 0.5 ml $litre^{-1}$ of spray solution. The foliar treatments were applied at 30, 45 and 60 days after sowing.

Table 3.5. Chemical composition of seaweed saps used

***Kappaphycus* sap**		***Gracilaria* sap**	
Nutrient	**Amount present**	**Nutrient**	**Amount present**
Moisture	94.38 g 100 ml^{-1}	Moisture	88.88 %
Protein	0.085 g 100 ml^{-1}	Crude protein	9.58 g 100 g^{-1}
Fat	0.0024 g 100 ml^{-1}	Crude lipid	2.00 g 100 g^{-1}
Crude fibre	0.01 g 100 ml^{-1}	Crude fibre	10.40 g 100 g^{-1}
Carbohydrate	1.800 g 100 ml^{-1}	Carbohydrate	45.92 %
Energy	7.54 Kcal 100 ml^{-1}	Saturated fatty acid	48.92 % of total fatty acids
Potassium	358.35 mg 100 ml^{-1}	Potassium	8633.00 mg 100 g^{-1}
Sodium	18.10 mg 100 ml^{-1}	Sodium	158.50 mg 100 g^{-1}
Magnesium	116.79 mg 100 ml^{-1}	Magnesium	549.50 mg 100 g^{-1}
Phosphorous	2.96 mg 100 ml^{-1}	Phosphorus	278.50 mg 100 g^{-1}
Calcium	32.49 mg 100 ml^{-1}	Calcium	32.49 mg 100 ml^{-1}
Iron	8.58 mg 100 ml^{-1}	Iron	67.35 mg 100 g^{-1}
Manganese	0.22 mg 100 ml^{-1}	Manganese	0.22 mg 100 ml^{-1}
Nickel	0.35 mg 100 ml^{-1}	Nickel	0.92 mg 100 g^{-1}
Copper	0.077 mg 100 ml^{-1}	Copper	0.20 mg 100 g^{-1}
Zinc	0.474 mg 100 ml^{-1}	Zinc	1.00 mg 100 g^{-1}
Chromium	3.50 mg 100 ml^{-1}	Chlorine	1170.00 mg 100 g^{-1}
Lead	0.51 mg 100 ml^{-1}	Lead	1.11 mg 100 g^{-1}
Indole acetic acid	23.36 mg L^{-1}	Cobalt	0.24 mg 100 g^{-1}
Gibberelin GA3	27.87 mg L^{-1}	Sulphate	106.20 mg 100 g^{-1}
Lead	1.11 mg 100 g^{-1}	Cadmium	0.14 mg 100 g^{-1}
Riboflavin	0.010 mg 100 ml^{-1}	Vitamin C	28.50 mg 100 g^{-1}
Iodine	160 mg 100 ml^{-1}	Total amino acids	889.78 mg g^{-1} of protein
Kinetin + Zeatin	31.91 mg L^{-1}		



(A) Main plot treatments		(B) Sub plot treatments	
Phosphorus levels P_1 = 20 kg P_2O_5 ha^{-1} P_2 = 40 kg P_2O_5 ha^{-1} P_3 = 60 kg P_2O_5 ha^{-1}	**Sulphur levels** S_0 = 00 kg S ha^{-1} S_1 = 20 kg S ha^{-1} S_2 = 40 kg S ha^{-1}	**Foliar sprays** F_0 = Control (Water spray) F_1 = *Kappaphycus* sap 10% F_2 = *Gracilaria* sap 10%	**Design** Split plot Plot size : 5.0 m x 3.6 m

Fig 3.2: Plan of Layout, 2012-13 and 2013-14

3.5.3 Seed and sowing

Most popular and recommended chickpea variety “Pratap channa-1” was sown in the experiment using seed rate of 80 kg ha^{-1}. The 10-12 cm deep furrows were opened with the help of kudali in each plot at row spacing of 30 cm. Fertilizers were placed in furrows as per treatment according to layout plan. After fertilizer placement the furrows were covered with soil at 2-3 cm height. The crop was sown in the same furrows where fertilizers were placed at depth of about 6-8 cm. During both the year sowing was done in dry condition followed by irrigation to secure proper germination. After sowing, the seeds were covered with soil droppings in order to ensure proper seed soil contact. Before sowing seeds were treated with fungicides Carbendazim at 2.0 g kg^{-1} seed to protect it from fungal diseases and then Chlorpyrifos at 4 ml kg^{-1} seed to protect the seed from termites. Thereafter, it was inoculated with *Rhizobium* culture.

3.5.4 Intercultural operations

Thinning of plants was done at 15 DAS by removing extra plants in order to maintain desired plant to plant spacing of 10 cm followed by one hand weeding at 35 DAS to provide effective control of weeds in chickpea crop.

3.5.5 Irrigation

First irrigation was given at the time of sowing. Second was given as per the crop requirement and to maintain the optimum moisture level in the field.

3.5.6 Harvesting and threshing

The crop was harvested at physiological maturity when plants turned golden yellow. The plants from border areas were harvested first, collected and removed from each plot. After this, crop in net plot was harvested, bundled and tagged separately. These bundles were brought to the threshing floor and left for sun drying for a period of 10 days. The dried bundles were weighed to record biological yield. After threshing, winnowing and cleaning was done and grains were weighed separately to record grain yield kg $plot^{-1}$ and converted respective observation to kg ha^{-1}. The grain and haulm samples from each experimental plot were collected for laboratory studies.

3.6 TREATMENT EVALUATION

3.6.1 Biometric studies

3.6.1.1 Growth parameters

a) **Plant height:** The height of five randomly selected plants from each plot was measured from ground surface to the tip of the main shoot at 30, 60 DAS and at harvest. The mean height was expressed in cm.

b) **Number of primary branches plant^{-1}:** The numbers of primary branches were counted from randomly selected five plants from each plot and average was recorded separately at 60 DAS and at harvest.

c) **Dry matter accumulation:** Five randomly selected plants from destructive sampling area in each plot at 30, 60, 90 DAS and at harvest (Physiological maturity) were collected. The sample plants were separated in to leaves, stem and reproductive parts and put into perforated paper bags separately. These samples were dried in sunlight for 2-3 days and then oven dried at 65º C for 72 hrs to obtain constant dry weight. Thereafter, the samples were weighed for estimating total dry matter accumulation (g plant^{-1}) and dry matter for leaves, stem and reproductive parts (g plant^{-1}) under each treatment at the above mentioned growth stages.

d) **Crop growth rate (g m^{-2} day^{-1}):** Crop growth rate (CGR) is the rate of dry matter production per unit ground area per unit time (Watson, 1952). It was calculated at 30-60 and 60-90 DAS by using the following formula

$$CGR = \frac{(W_2 - W_1)}{(t_2 - t_1)} \times \frac{1}{A}$$

Where, W_1 = Dry weight of the plants (g m^{-2}) at time t_1

W_2 = Dry weight of the plants (g m^{-2}) at time t_2

t_1-t_2 = Time interval in days

A = Unit land area in (m^2)

e) **Absolute growth rate (g plant^{-1} day^{-1}):** Absolute growth rate (AGR) expresses the dry weight increase per unit time and was calculated at 30-60 and 60-90 DAS by using the following formula,

$$AGR = \frac{W_2 - W_1}{t_2 - t_1}$$

Where, W_2 and W_1 are the total dry weights per plant in gram at t_2 and t_1 times in days, respectively.

f) **Relative growth rate (g g^{-1} day^{-1}):** Relative growth rate (RGR) is rate of increase in dry weight per unit dry weight already present. Relative growth rate at various stages was calculated at 30-60 and 60-90 DAS as suggested by Radford (1967).

$$RGR = \frac{\log_e W_2 - \log_e W_1}{t_2 - t_1}$$

Where, W_1 = Dry weight of plants (g) at time t_1

W_2 = Dry weight of plants (g) at time t_2

g) **Biomass duration (g days):** Biomass duration (BMD) is calculated at 30-60 and 60-90 DAS by using the following formula

$$BMD = \frac{TDMi + TDM(i+1)}{2} \times (t_2 - t_1)$$

Where,

TDMi = TDM at i^{th} stage

TDM (i+1) = TDM at $(a+1)^{th}$ stage

$(t_2 - t_1)$ = Time interval between i^{th} stage and $(i+1)^{th}$ stage (days)

3.6.1.2 Yield components

a) **Number of pods plant^{-1}:** The pods of five plants counted and the mean value was recorded as number of pods per plant from each treatment.

b) **Number of grains pod^{-1}:** The grains of five pods from each plant counted and the mean value was recorded as number of grains per pods from each treatment.

c) **Number of grains plant^{-1}:** The grains of five plants counted and the mean value was recorded as number of grains per plant from each treatment.

d) **Grain yield plant^{-1}:** The randomly selected five plants of net plot area taken for the weight of the grains per plant. The grains were weighed and average weight of grains per plant was computed and expressed as weight of grains per plant (g).

e) **100-grain weight:** The sun dried random grain sample from the yield of net plot was taken out and hundred grains were counted weighed for recording 100-grain weight in g.

3.6.1.3 Yield and harvest index

a) **Biological yield:** The weight of thoroughly sun-dried plants of net plot along with pods was recorded and expressed as biological yield in kg ha^{-1}.

b) **Grain yield:** After threshing and winnowing, grain yield plot^{-1} was weighed and expressed in term of kg ha^{-1}.

c) **Haulm yield:** Haulm yield was obtained by subtracting the grain yield per plot from the respective biological yield per plot and finally expressed in terms of haulm yield in kg ha^{-1}**.**

d) **Harvest index:** The harvest index was obtained by dividing the economic yield (grain yield) by total biological yield and expressed as per cent (Donald and Hamblin, 1976).

$$\text{Harvest index (\%)} = \frac{\text{Economic yield (kg ha}^{-1}\text{)}}{\text{Biological yield (kg ha}^{-1}\text{)}} \times 100$$

3.6.1.4 Quality parameters

The chickpea grain samples collected at harvest from each plot were oven dried at 65º C for 72 hrs. The dried samples were finely ground and used for estimation of protein and amino acids *viz*., methionine, cysteine and cystine content as per method furnished in Table 3.6.

Table 3.6 Methods used for quality analysis

S. No.	Characters	Reference
1.	Protein	A.O.A.C. (2002)
2.	Methionine	Thimmaiah (1999)
3.	Cysteine	A.O.A.C. (2002)
4.	Cystine	A.O.A.C. (2002)

Preparations of plant samples for chemical analysis

The experimental plant samples were prepared as per procedure detailed below-

(i) Plant samples for elemental composition

Selected plants randomly from individual plots. These representative samples were washed in running tap water, 0.01 N HCL and thoroughly rinsed with distilled water in succession. These were then dried in an air forced oven at 65^0C to constant weight. Dried samples were finally ground in Wiley steel grinding mill provided with 20 mesh sieve, avoiding any metallic contamination.

(ii) Preparation of leaf for biochemical analysis

At 60 days after sowing, young uppermost leaves of plants were picked up, placed in polythene bags and immediately brought to the laboratory. They were thoroughly washed in running tap water, 0.1 N HCL and then distilled water in succession. Excess surface water was removed by putting in between folds of blotting papers. Leaves were chopped in to five pieces with scissors and made in to homogeneous samples before processing them for chlorophyll content.

3.6.2 Biochemical analysis

a) Estimation of chlorophyll content of leaves at 60 DAS: Chlorophyll content of fresh leaf samples was determined by using the colorimetric method at 60 DAS of crop (Arnon, 1949). The total chlorophyll content was determined by following formula:

$$\text{Total chlorophyll content (mg g}^{-1}\text{ fresh weight of leaf)} = 20.2\,(A645) + 8.02\,(A663) \times \frac{V}{1000 \times W\,(g)}$$

Where:-

V = volume of extract (ml)

W = weight of leaf sample (g)

b) Estimation of N, P, K and S content of leaves at 60 DAS and plant (grain and haulm) at harvest: The chickpea leaves collected at 60 DAS and plant samples prepared at harvest for analysis as per procedure described above. The samples were used for determination of N, P, K and S content as per method furnished in Table 3.7.

d) Nutrient (N, P, K and S) uptake

N, P, K and S uptake were computed from the data of N, P, K and S content of grain and haulm and grain and haulm yield using the following formula:

$$\text{Nutrient uptake (kg ha}^{-1}\text{)} = \frac{\text{Nutrient content (\%) in grain/haulm x grain/haulm yield (kg ha}^{-1}\text{)}}{100}$$

3.6.3 Soil analysis

Soil samples from 0-15 cm depth from three spots per plot were drawn before sowing and at harvest of the crop. These samples were mixed and composite samples were thus prepared for all experimental plots individually. Such samples after proper drying in shade were processed for available nitrogen, phosphorus and potassium contents as per the procedure referred in Table 3.7.

Table 3.7 Methods used for plant and soil analysis

S.N.	Properties	Procedure	Reference
A	**Plant analysis**		
1.	Digestion of plant sample	Wet digestion of plant samples with H_2SO_4 and H_2O_2 were carried out for determination of nitrogen content.	Jackson (1973)
1.1	Nitrogen	Colorimetric or spectrophotometer method using Nesseler's reagent.	Snell and Snell (1949)
2.	Digestion of plant sample	Wet digestion with di-acid mixture HNO_3 $HClO_4$ (9:4).	Johnson and Ulrich (1959)
2.1	Phosphorus	Vanado-molybdo- phosphoric acid yellow colour method	Jackson (1973)
2.2	Potassium	Flame Photometer method	Jackson (1973)
3.1	Sulphur	Turbidimetrically colorimetric method	Tabatabai and Bremner (1970)
B.	**Soil analysis**		
1.	Available N (kg ha^{-1})	Alkaline permanganate method	Subbiah and Asija (1956)
2.	Available P (kg ha^{-1})	Olsen's P, 0.5 M $NaHCO_3$, pH 8.5 extractable P method	Olsen *et al.*(1954)
3.	Available K (kg ha^{-1})	Neutral ammonium acetate extractable K and Flame photometry	Richard (1954)
4.	Available S (mg kg^{-1})	Calcium chloride extractable S method	Williams and Steinberg (1959)

3.6.4 Economics of the experimental treatments

a) Net returns (₹ ha^{-1})

To find out the most profitable treatment, economics of different treatments was worked out in terms of net monetary returns (₹ ha^{-1}) by subtracting the cost of treatment and the cost of cultivation from gross income obtained. Cost of cultivation and net profit were calculated on the basis of prevailing prices of produce and inputs (Appendix XXII).

(ii) Benefit-cost ratio

This was calculated by dividing net returns with cost of cultivation for each treatment to see the economic viability of treatments.

3.7 Statistical analysis

3.7.1 Analysis of variance and test of significance

In order to test the significance of variation in experimental data obtain for various treatment effects, data were statistically analysed as described by Panse and Sukhatme (1989). The critical difference was calculated to assess the significance of treatment mean wherever, the "F" test was significant at 5 per cent level of significance. In order to elucidate the nature and the magnitude of effects, summary tables along with S.Em. ± and C.D. at 5 per cent level are embodied in the next chapter " Experimental results" and their analysis of variance are given in the appendices (I-XXI) at the end. The homogeneity of error variances is tested using Bartlett (1947) chi-square test (x^2). The error variances in the experiment were found homogeneous hence, pooled analysis were carried out for the said experimental design.

3.7.2 Correlation and regression studies

Correlation studies were carried out with a view to determine interrelationship between various characters as described by Panse and Sukhatme (1989). Regression equations for the characters indicating significant correlation were also worked out and presented at appropriate places.