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Effect of liquid fermented organic manure (Herbal kunapajala) concoctions and their doses on chlorophyll content of mustard crop at Pantnagar, India

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Abstract
In present agricultural practices, farmers are mainly dependent of external inputs which causes serious damage to our environment. With this theme, present investigation was focused on ancient (or) traditional practices that enhancing the growth and development of crop without hinder the environment by use locally available resources. The study was conducted during *rabi* 2020-2021 at D7 Block of Norman E. Borlaug Crop Research Centre, G. B. Pant University of Agriculture and Technology, Udham Singh Nagar, Uttarakhand, India. The design used for experimentation was factorial randomized block design (FRBD) with additional treatment. In factor A, three herbal kunapajala concoctions (Nettle based herbal kunapajala (KJ_1), General weeds based herbal kunapajala (KJ_2) and integrated based herbal kunapajala (KJ_3)) and in factor B four doses (500 l/ha (D_1), 1000 l/ha (D_2), 1500 l/ha (D_3) and 2000 l/ha (D_4)) with control as additional treatment (N_1). Among herbal kunapajala concoction treatments, significant difference was noticed at 60 DAS and 90 DAS. The higher chlorophyll a content at 60 and 90 DAS was observed in KJ_1 (0.72 and 0.64 mg(g/FM)) as compared to other treatments. Similarly, total chlorophyll content and carotenoid were significant at 60 DAS (1.88 and 0.64 (mg(g/FM), respectively). D_4 recorded significantly superior values of chlorophyll a, total chlorophyll and carotenoid at 60 DAS under doses treatments (0.71, 1.85 and 0.74 (mg(g/FM), respectively). The interaction effect was non-significant in all the parameters and crop growth stages. Among control versus rest treatment, control recorded significant difference of chlorophyll a (60 DAS), b (60 DAS), total chlorophyll (60 and 90 DAS) and carotenoid (60 DAS) as compared to rest treatment. Whereas, rest treatment recorded significantly superior chlorophyll a at 60 DAS. Based on the results, application of nettle based herbal kunapajala with 2000 l/ha will give higher chlorophyll content in mustard crop.

Keywords: Liquid fermented organic manure, herbal kunapajala, chlorophyll content, mustard, recommended dose of nutrient

Introduction
The present aeon of Indian agriculture is facing the problem of downward spiralling of natural resources due to continuous use of synthetic fertilizers particularly after green revolution. Green revolution was introduced with ground to make food needs of population self-sufficient by boosting crop productivity as only focus. Due to this, unintentionally inappropriate application rates of fertilizer practices were adopted widely by the farmers. Further, with fertilizer subsidy scheme (1977), consumption of synthetic fertilizer has been increasing from past four decades. The result was increase in food grain production but at the cost of deterioration of natural resources. Thus, farming community is forced to vortex of dire necessity to twin focus on maximisation of crop production sustainably and preservation of natural resources. In this context, organic manure that has inherent capacity not only to supply essential micro and macronutrients for balanced and timely contribution on growth and development of plant but also for enhancing soil health will be the need of hour.

The above lines are more related to herbal kunapajala, fermented liquid organic manure mentioned in Vrikshayurveda written by Surapala around 1000 AD and in Lokopakara written by Chavundaraya in 1025 AD (Chakraborty *et al.* 2019) [1]. Herbal kunapajala, an alternate nutrient source against chemical fertilizer and it is highly effective in crop growth and development. Herbal kunapajala can be used as nutrient supplement at any growth stage of crop plant. The effectiveness of herbal kunapajala is due to the breakdown of complex compounds in to lower molecular weight compounds during the fermentation of ingredients and this make available nutrients to the plants (Neff *et al.*, 2003) [6].

Liquid organic manure plays important role in enhancing immunity (Sreenivasa *et al*., 2010) [11], vigour and quality to plant system. Herbal kunapajala contains higher amount of macro and micronutrients and enhances soil fertility. Due to increase in availability of nutrients especially nitrogen and magnesium in soil, easily absorbed by the plant. Chlorophyll content is made by porphyrin ring, contains nitrogen and magnesium as major nutrients in structure. Whereas, chlorophyll content helps for photosynthetic activity in plants and produce assimilates. Due to increase in chlorophyll content per unit area, enhances in assimilates production and ultimately increase in dry matter. Additionally, it also supplies growth promoting hormones *viz.*, IAA and GA_3 (Palekar, 2006; Natarajan, 2007; Sreenivasa *et al*., 2010) [7, 5, 11]. The aim of the study is knowing the best management practices under organic agriculture in comparison with conventional farming on chlorophyll content of mustard crop.

Materials and Method

A field experiment was conducted during *rabi* season 2020-21 at D7 Block of Norman E. Borlaug Crop Research Centre, G. B. Pant University of Agriculture and Technology, Udham Singh Nagar, Uttarakhand, India. The experimental plot is located at latitude of 29^0 01' N and longitude of 79^0 48' E and at an altitude of 243.83 meters above mean sea level (MSL). The mean average rainfall of the experiment site was 1450 mm. The soil of the experimental site belongs to order Mollisol. The experiment was laid out in factorial randomized block design (FRBD) with additional treatment and replicated thrice (Rangaswamy, 1995) [9]. In factor A, three herbal kunapajala concoctions (Nettle based herbal kunapajala (KJ_1), General weeds based herbal kunapajala (KJ_2) and integrated based herbal kunapajala (KJ_3)) and in factor B four doses (500 l/ha (D_1), 1000 l/ha (D_2), 1500 l/ha (D_3) and 2000 l/ha (D_4)) with control as additional treatment (N_1). The experiment unit size was 6.0 m × 2.5 m and with spacing 30 cm × 10 cm. Kranti variety was selected for experimentation, released by GBPUAT, Pantnagar in 1988 at national level. The tillage operation was done with tractor mounted disc harrow, rotavator into the field to acquire fine seed bed for good germination of mustard seed. Thereafter, field was levelled with pata. The bunds were made with small bund maker at borders and in between the plots man made bunds are formed.

A seed rate of 5 kg/ha was used for sowing mustard crop. The seed treatment was done with 3 different types of herbal kunapajala separately according to the treatment and requirement of seed per plot. The seeds were soaked for 5-10 min in herbal kunapajala and later dried for overnight until seed were fully dried. In case, 100% RDF treatment seeds were treated with gaucho @120 ml/100 kg of seed for controlling seed borne diseases. The herbal kunapajala was prepared by addition of cow dung and cow urine (20 kg and 20 l) in a plastic drum with a capacity of 200 litres. The sprouted urd (2 kg), mustard cake (2 kg) and crushed jaggery (2 kg) were added in a plastic drum. Thereafter, 20 litres water was added for proper mixture of ingredients. The fresh nettle plants (chopped into small pieces) were weighed 20 kgs and added into the drum. Paddy husk was boiled 2 days before the herbal kunapajala preparation for 15 – 20 minutes in water and filtered the contents. The filtered contents were added into drum. The milk and butter milk (1 litre each) were added in to the drum. After addition of all the ingredients once stir with wooden stick and later water was added up to the opening of lid. The lid was closed after preparation. Every day morning and evening stirring should be done with the wooden stick for 10 min. Once the fermentation process starts, we can see the bubbles on the top of the solution. After 20-25 days the bubbles stop to appear and we can notice that the herbal kunapajala is ready for application in field. The solution was filtered with mesh to remove the residue and herbal kunapajala solution was stored for further use. In case of general herbal kunapajala preparation, the process and ingredients was same as nettle based herbal kunapajala preparation except addition of nettle plant we use 2 kg each of freshy chopped leaves and plants of general weeds *viz.*, neem (*Azadirachta indica*), wild jasmine (*Clerodendrum* sps.), beal (*Aegle marmelos*), datura (*Datura* sps.), lantana (*Lantana camara*), mango (*Mangifera indica*), guava (*Psidium guajava*), calotropis (*Calotropis* sps.), castor (*Ricinus communis*) and billy goat weed (*Ageratum conyzoides*), respectively. Whereas, integrated herbal kunapajala was prepared by mixture of half the quantity of nettle plants and general weeds and remaining process was same. The herbal kunapajala and their doses were applied according to treatment wise and in control treatment 100-20-60-20 kg N - P_2O_5 - K_2O-S/ha was applied.

The total chlorophyll content of leaves will be recorded on 30, 60 and 90 DAS by using SPAD chlorophyll meter and NDVI. In case of, chlorophyll a, b and carotenoids content will be measured at 30, 60 and 90 DAS in fresh leaves by using Dimethyl Sulfoxide (DMSO) method described by Hiscox and Israelstam (1979) [4].

$$NDVI = \frac{\text{NIR reflectance} - \text{Red reflectance}}{\text{NIR reflectance} + \text{Red reflectance}}$$

$$\text{Chlorophyll a} = (12.47 \times A_{665}) - (3.62 \times A_{649})$$

$$\text{Chlorophyll b} = (25.06 \times A_{649}) - (6.50 \times A_{665})$$

$$\text{Total chlorophyll} = \text{chl a} + \text{chl b}$$

$$\text{Carotenoids} = (1000A_{480} - 1.29 \text{ Chl a} - 53.78 \text{ chl b}) / 220$$

The experimental data obtained during course of investigation will be subjected to statistical analysis by using analysis of variance (ANOVA) technique prescribed for factorial randomized block design with additional treatment (Rangaswamy, 1995) [9]. F test will be used to test the significance of overall variation among treatments and conclusion will be drawn at 5 per cent probability level. The critical difference (CD) will be computed to test the significant different between the treatments.

Results and Discussion

The chlorophyll a content as influenced by different treatments at various intervals of crop growth are presented in Table 1. Among herbal kunapajala concoction treatments, significant difference was noticed at 60 DAS and 90 DAS. The higher chlorophyll a content at 60 and 90 DAS was observed in KJ_1 (0.72 and 0.64 (mg(g/FM))) as compared to other treatments. Whereas, at 30 DAS chlorophyll a content was maximum in KJ_3. Among doses treatment, significantly superior chlorophyll a content was observed at 60 DAS. D_4 recorded higher values over other treatments under doses treatments (0.54, 0.71 and 0.65 (mg(g/FM)). Across

interaction treatments, non-significant results were observed. Among control versus rest treatments, significant effect was observed at 30 and 60 DAS. The higher values were noticed in control treatment as compared to rest treatment except 30 DAS.

The chlorophyll b content was non-significant under herbal kunapajala concoction treatments. Whereas, KJ_1 recorded higher chlorophyll b content (0.63, 1.16 and 0.81 (mg(g/FM)) at respective crop growth stages as compared to other treatments. Similarly, doses and interaction treatments also showed non-significant difference at different crop growth stages. At 30 DAS, D_1 (0.64 (mg(g/FM)) recorded maximum values as compared to other treatments. Similarly, at 60 and 90 DAS D_4 and D_3 (1.15 and 0.85 (mg(g/FM), respectively) recorded higher values. Across control versus rest treatments, non-significant effect was noticed but control treatment recorded higher chlorophyll b content (0.62, 1.24 and 0.93 (mg(g/FM)) as compared to rest treatment.

Across herbal kunapajala concoction treatment, total chlorophyll content showed non-significant effect except at 60 DAS. KJ_1 observed higher values at 60 and 90 DAS (1.88 and 1.44 (mg(g/FM)) as compared to other treatments. Similarly at 30 DAS KJ_3 recorded higher total chlorophyll content. Under doses treatment, significant effect was noticed at 60 DAS. Whereas, D_4 recorded higher values at 60 DAS (1.85 (mg(g/FM)) as compared to other treatments. Non-significant effect was observed under interaction treatment. Across control versus rest treatment, significant effect was produced during 60, 90 DAS. Control treatment recorded maximum total chlorophyll content (2.08 and 1.57 (mg(g/FM)) except 30 DAS over rest treatment.

The carotenoid content as influenced by different treatments at various intervals of crop growth stages are presented in Table 2. Among herbal kunapajala concoction treatment, non-significant difference between the treatments were noticed except 60 DAS. The carotenoid content was recorded maximum in KJ_1 treatment (0.53, 0.64 and 0.55 (mg(g/FM)) as compared to other treatments. Under doses treatment, significant effect was noticed in 60 DAS. Whereas, D_4 recorded higher carotenoid content (0.58, 0.74 and 0.58 (mg(g/FM)). The interaction effect was non-significant. Among control versus rest treatment, significant effect was observed during 60 DAS, higher carotenoid content was recorded with control treatment (0.56, 1.10 and 0.61 (mg(g/FM)) as compared to rest treatment.

Data related to SPAD and NDVI values as influenced by different treatments are illustrated in Fig 1. Across herbal kunapajala concoctions, SPAD values showed non-significant effect at respective crop growth stages. The higher SPAD values were observed in KJ_1 (332.87, 437.10 and 293.77) as compared to other treatments. Similarly, doses treatment also showed non-significant but D_1 recorder maximum values at 30, 90 DAS. However, at 60 DAS higher SPAD values was observed with D_2 as compared to other treatments. The interaction effect of treatments was recorded non-significant. Under control versus rest treatments, non-significant effect was noticed but higher SPAD values were observed in control treatment as compared to rest treatment. Non-significant effect of NDVI values was observed under herbal kunapajala concoctions. KJ_1 recorded maximum NDVI values (0.43, 0.57, 0.36as compared to other treatments. Across doses treatment, non-significant effect was produced but higher NDVI values were noticed under D_4 treatment (0.42 and 0.37) except 60 DAS. The interaction treatments effect showed non-significant during both years. Under control versus rest treatments, non-significant effect was observed, higher NDVI values were recorded in control treatments over rest treatment.

Chlorophyll content is a good index of photosynthetic activity, stress and plant health. Nitrogen is a constituent of leaf chlorophyll content and in each chlorophyll structure four nitrogen atoms are bonded. The higher nitrogen content in nettle based herbal kunapajala might be the reason for enhancing total chlorophyll, carotenoid and chlorophyll a and b content in mustard plant. The nettle based herbal kunapajala was applied as soil and foliar application which might enhanced the availability of nutrients to plants. As foliar application of herbal kunapajala has directly absorbed by the leaf through stomatal opening. Leaf area also plays a major role in chlorophyll content per unit leaf area. Deshmukh *et al.* (2012) [2] noticed increase in no of leaves per plant with kunapajala treatment as compared to conventional farming and organic farming (9:1 ratio of soil and vermicompost mixture). Further, leaf area index, total chlorophyll, carotenoids and xanthophylls were also observed maximum under kunapajala treatment over other treatments. Similarly, higher pigment content (chl a, chl b and carotenoid) at seedling, flowering and yielding stage of tomato crop was noticed with application of 3% panchagavya spray over control and other panchagavya concentrations (Rakesh *et al.*, 2017) [8]. According to Somasundaram *et al.* (2007) [10], reported bigger leaves and denser canopy of plants sprayed with panchagavya. Similar findings were observed in *Arachis hypogaea* (Subramaniyan, 2005) [12] and *Vigna radiate, Vigna mungo* and *Oryza sativa* (Tharmaraj, 2011) [13]. SPAD and NDVI values are indirect measurement of total chlorophyll content in plants. It also a good indicator for plant health. Similar findings were noticed by Gayathri and Aiswariya (2020); Upadhyay *et al.* (2019) [3, 14]. In case of doses, D_4 recorded maximum chlorophyll values in all the parameters. This might be due to the application of higher dose of herbal kunapajala which contains more amount of plant available form of macro and micronutrients especially nitrogen content.

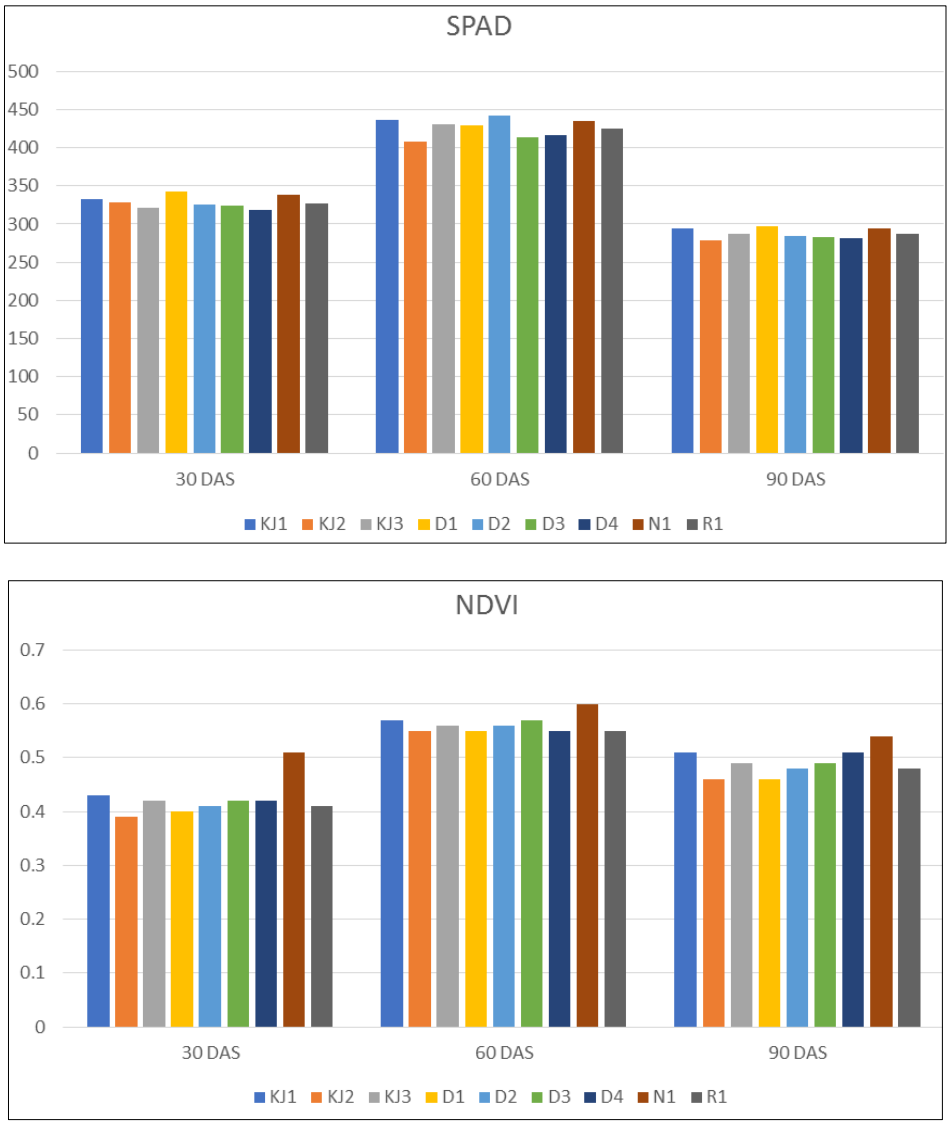

Fig 1: Effect of herbal kunapajala concoctions and their doses on SPAD and NDVI values of Indian mustard at different growth stages

Table 1: Effect of herbal kunapajala concoctions and their doses on Chlorophyll a and b content of Indian mustard at different growth stages

Treatment	Chlorophyll a content (mg(g/FM))			Chlorophyll b content (mg(g/FM))		
	2020-2021			2020-2021		
	30 DAS	60 DAS	90 DAS	30 DAS	60 DAS	90 DAS
Herbal kunapajala concoctions						
KJ_1	0.53	0.72	0.64	0.63	1.16	0.81
KJ_2	0.53	0.58	0.59	0.60	1.09	0.81
KJ_3	0.55	0.64	0.63	0.62	1.12	0.79
S.Em±	0.005	0.020	0.013	0.013	0.027	0.019
CD (p=0.05)	NS	0.05	0.03	NS	NS	NS
Doses						
D_1	0.53	0.59	0.60	0.64	1.06	0.77
D_2	0.54	0.62	0.61	0.60	1.15	0.80
D_3	0.53	0.67	0.62	0.61	1.14	0.85
D_4	0.54	0.71	0.65	0.62	1.15	0.78
S.Em±	0.006	0.023	0.015	0.015	0.031	0.022
CD (p=0.05)	NS	0.06	NS	NS	NS	NS
Interaction						
S.Em±	0.010	0.039	0.025	0.025	0.053	0.038
CD (p=0.05)	NS	NS	NS	NS	NS	NS
Control vs rest						
Control	0.50	0.83	0.64	0.62	1.24	0.93
Rest	0.53	0.64	0.62	0.61	1.12	0.80
S.Em±	0.011	0.041	0.027	0.027	0.056	0.040
CD (p=0.05)	0.03	0.11	NS	NS	0.16	NS

Table 2: Effect of herbal kunapajala concoctions and their doses on total Chlorophyll and carotenoid content of Indian mustard at different growth stages

Treatment	Total chlorophyll content (mg(g/FM))			Carotenoid content (mg(g/FM)		
	2020-2021			2020-2021		
	30 DAS	60 DAS	90 DAS	30 DAS	60 DAS	90 DAS
Herbal kunapajala concoctions						
KJ_1	1.16	1.88	1.44	0.53	0.64	0.55
KJ_2	1.13	1.67	1.40	0.52	0.52	0.54
KJ_3	1.17	1.75	1.42	0.52	0.61	0.55
S.Em±	0.013	0.030	0.027	0.028	0.027	0.038
CD (p=0.05)	NS	0.08	NS	NS	0.07	NS
Doses						
D_1	1.17	1.65	1.38	0.47	0.48	0.47
D_2	1.14	1.76	1.40	0.56	0.50	0.57
D_3	1.14	1.80	1.48	0.48	0.65	0.57
D_4	1.16	1.85	1.43	0.58	0.74	0.58
S.Em±	0.015	0.035	0.031	0.032	0.031	0.044
CD (p=0.05)	NS	0.10	NS	NS	0.09	NS
Interaction						
S.Em±	0.026	0.060	0.053	0.055	0.053	0.076
CD (p=0.05)	NS	NS	NS	NS	NS	NS
Control vs rest						
Control	1.13	2.08	1.57	0.56	1.10	0.61
Rest	1.15	1.76	1.42	0.52	0.59	0.54
S.Em±	0.028	0.063	0.056	0.058	0.055	0.080
CD (p=0.05)	NS	0.18	0.16	NS	0.16	NS

Conclusion

On the basis of experimentation, nettle based herbal kunapajala and D_4 (2000 l/ha) gave higher chlorophyll a, b, total chlorophyll, carotenoid, SPAD and NDVI values. Control treatment noticed better pigment content. Further, application of liquid fermented organic manures may increase the nutrient content in soil and make easily available to plant. It also indirectly enhances the plant health, productivity with sustainability.

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