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Influence of Pranic Agriculture in Enhancing Growth and Yield of Chilli (*Capsicum annuum* L.) and its Genetic Analysis

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ABSTRACT

Pranic Agriculture (PA) is an emerging supplementary farming system applied to the seeds, soil, and seedlings to improve plant growth and yield by improving the quality and quantity of Prana using Pranic Healing techniques. Chilli is the major spice crop of India. There is a constant demand for chilli in the local and international markets; necessitating its increased production. A field study (0.2 h) was carried out to understand the influence of Pranic agriculture treatment on chilli plant growth parameters, flowering, yield traits and molecular changes at the DNA level by Random Amplified Polymorphic DNA (RAPD) marker analysis. Plant height was significantly (p<0.05) higher in pranic treatment by 21% than in control. Leaf Chlorophyll content and root length were increased by 13% and 44% respectively in pranic treated chilli than control. Pranic treatment showed significantly higher fruit weight (6.47 g), the number of seeds per fruit (64.5), and yield per acre (14.25 t) as compared to control (5.56g, 51.1 and 12.21t) respectively. RAPD marker analysis recorded an average polymorphism of 24% between pranic and control. Pranic treatment was found effective in increasing the growth and yield of chilli. Pranic Agriculture would be a supportive farming system for attaining sustainable and eco-friendly agriculture. **Keywords:** Chlorophyll, Crop improvement, Prana; Pranic Healing, Sustainable agriculture

INTRODUCTION

Chilli (*Capsicum annuum* L.) is one of the most valuable spice crops for small and marginal farmers in Asia, Africa, and South America and it is grown throughout the world. India is the largest producer of Chilli followed by China, Indonesia, and Korea and stands first in consumption and export of chillies. Both green and red chillies are the most important ingredients of the Indian daily diet. Almost 400 types of chillies are grown throughout the world and many of the hottest varieties are grown in India. Major chilli production is from Andhra Pradesh, Maharashtra, Karnataka, Gujarat, Tamil Nadu, and Orissa in India (Mehta, 2017). The extracts of Chilli are used to alleviate the pain of arthritis, headaches, burns, and neuralgia (Pundir *et al.*, 2016; Saleh *et al.*, 2016). There is tremendous scope for export of chillies as a spice and still, the chilli production in India is not optimum to meet the demand. So, increasing yield potential and quality is a necessary approach to satisfy the increasing demand in both local and international markets. One of the major objectives in chilli is to obtain higher yields and to achieve this yield-related traits like the number of fruits per plant, number of seeds per fruits, fruit girth, fruit length and fruit weight need to be improved (Balraj *et al.*, 2018; Visalakshi and Pandiyan, 2018).

Pranic agriculture (PA) is a science and art of cultivation of crops by application of '*Prana*' to the plant through pranic healing techniques. According to Master Choa Kok Sui, Pranic treatment uses prana to heal the bioplasmic body (Sui, 2015). Prana or life force is a bioplasmic body or "*Aura*" which surrounds all the living organisms on earth like man, animals, and plants. The word bioplasmic refers to 'bio' means life and plasma, which is the fourth state of matter. Scientists with the help of Kirlian's photography (Kirlian, 1949) have rediscovered the bioplasmic body that keeps the living organism healthy and alive. Plants absorb prana from sun, air, soil, and water. Pranic treatment is given to the plants by a trained Pranic healer (Sui, 2015).

Pranic treatment on Drumstick and Papaya seedlings showed significantly higher mean germination rate, the number of leaves, shoot length, and seedling vigor when compared to control (Prasad *et al.*, 2019; Prasad and Jois, 2020). By applying pranic treatment, morphological traits like plant height and plant spread were increased respectively by 18.5% and 12% in tomatoes (Jois *et al.*, 2016) and plant height and stem diameter by 97% and 32% respectively in cucumber (Jois *et al.*, 2017) against its untreated group. Flowering traits like days to flower initiation, flowering to fruit set were reduced by 2.2% and 3.2% in pole beans (Yathindra *et al.*, 2017) and days to 50% flowering was reduced by 8.8% in European cucumber (Yathindra *et al.*, 2017) as compared to control. Similarly, the yield was also increased by pranic energy application in Tomato by 31%, Cucumber by 20.8%, Pole beans by 10.7%, and European cucumber by 18%, against its control group. Pranic treated Brinjal fruits retained better storage characteristics like firmness, non-decay, colour, gloss, and non-shrivelling, for a longer time as compared to control groups (Jois *et al.*, 2019). Significant improvements observed in the previous studies, highlights the importance of Pranic agriculture in near future as a supplementary and complementary eco-friendly farming system supportive to other existing agricultural practices and technologies. In the present study, the influence of Pranic agriculture method is adopted in chilli crop towards sustainable agriculture. Differences observed for morphological traits were correlated with a genetic polymorphism at the DNA level using RAPD molecular marker analysis.

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MATERIAL AND METHODS

Material and Treatment Method:

Pranic agriculture study was carried out at the College of Horticulture, Mysore in 2019 under field conditions during *Kharif* season. The study coordinates were 12.38015N, 76.52426E. The energy in the form of prana is projected to seed, cocopeat, soil, and crop by using a standard protocol (Sui, 2015). Pranic energy was applied by a trained pranic healer for 15 min for 6 times with an interval of twice a week. The seed, cocopeat, soil, and crop with no treatment was referred to as the control group. Chilli seeds were treated with pranic energy and sown in the trays filled with pranic energy treated cocopeat and another set without treatment. Both sets were grown in nursery for up to 25 days and later they were transplanted to their respective field plots for further studies. While seeds were establishing in the nursery, simultaneously land was treated with pranic energy and another set of land is maintained as control. Transplantation was done in their respective treated plots in 3 replicates (0.2 ha each) with a spacing of 75 cm between the row and 45 cm between the plants. Experiments was conducted in total 6 randomised blocks for 2 treatments (control and pranic). Both the study plots consist of red loam soil and the same environmental conditions. Drip irrigation was adopted for the crop. The recommended dose of FYM (20 tons) is given before transplanting and NPK fertiliser @ 150:75:75 kg/ha was given in two split doses. Weeding and other necessary cultural operations are taken care as required (Fig. 1).



Fig. 1. Experimental view of chilli plots (Pranic and control)

Data collection:

Healthy and uniform 25 days old chilli seedlings were transplanted to the main field from nursery and were allowed for establishment. Plant growth and development was assessed by the measurement of plant height and stem girth at vegetative growth stages of crop. Morphological growth traits like plant height (cm), stem girth (cm) was recorded at 75 days after transplant in both pranic and control plots. The height of the plant was measured using a measuring tape from ground level to the shoot tip. Similarly, stem girth is the circumference of the stem measured using the measuring tape. Flowering traits like days taken for first flowering, 50% flowering, and first harvest were recorded when we observe flower and fruit emergence. Yield and its related traits like root length (cm), fruit length(cm), fruit weight (g), number of fruits per plant, fruit girth (cm), number of seeds per fruits, and fruit yield per acre (t) were recorded after harvest. Taproot length from the ground level up to the end of the root was measured using a measuring tape.

Measurement of Total Chlorophyll Content:

Total Chlorophyll content of chilli leaves was estimated using Dimethyl sulphoxide (DMSO) (Shaof and Lium, 1976).

Molecular Analysis:

DNA was isolated from 0.5 g of 2nd or 3rd young fresh leaf from top in control and pranic treated groups by standard method with slight modifications (Rezadost *et al.*, 2016). For individual treatment, 9 leaf samples were collected randomly from three blocks and DNA is extracted individually and then bulked into one sample. Further, the DNA quality was observed on 0.8% agarose gel, and purity was assessed by A_{260}/A_{280} absorbance ratio using a nano drop. The plant genomic DNA was subjected to PCR using arbitrary universal oligonucleotide primers for tracing genetic polymorphism. Three decamer primers GE2, GE3 (GE healthcare), and OPL-12 (Operon Technologies Inc., Alameda CA. USA) were used for RAPD analysis. RAPD marker analysis was carried out to know the effect of pranic treatment on DNA characteristics of plants. The reaction mixture final volume was 25 µl containing the following components; DNA template: 5 µl (10 ng), primer: 2 µl (20 pmol), dNTPs: 5 µl (0.2 mM), *Taq* DNA polymerase: 0.5 U, PCR buffer: 2.5 µl, MgCl₂: 1.5 mM and H₂O: 8.5 µl. The PCR programme was executed with one cycle of denaturation at 95°C/3min, followed by 35 cycles of

 $95^{\circ}C/45$ sec, $36-40^{\circ}C/1$ min, $72^{\circ}C/1$ min and a final extension of $72^{\circ}C/5$ min. Amplified PCR products along with a 10 kb DNA ladder were separated by 1.8% (w/v) agarose gel and was stained with ethidium bromide and documented using a gel doc system. The percentage of polymorphism was calculated using the equation. **Percent polymorphism= A/B×100**

Where, A = number of polymorphic bands; and B = the total number of scorable bands.

Data Analysis:

The morphological and yield data were analysed for 10 samples from each treatment (pranic and control). Analysis was carried out by Paired *t*-test using Microsoft Excel and expressed as Mean \pm SD and the level of significance were expressed at p=0.05 (Kim, 2015).

RESULTS

Morphological and Yield Traits:

Plant height (cm) was significantly higher in pranic treatment by 21.91% when compared to control (**Table 1**). Stem girth (cm) was higher by 8% in pranic treatment when compared to control, but the results were non-significant. Chlorophyll content (mg/g) is one of the major pigments involved in the photosynthetic activity and was improved up to 13.64% by pranic treatment against control (Table 01). Treatment showed 4.25 mg/g chlorophyll content compared to the control of 3.74 mg/g. The root length of pranic treatment was significantly higher than control which accounts for 44.3% improvement.

Table 1. Influence of pranic treatment on chlorophyll content, plant growth, flowering and yield of chilli (N=10)

Tueite	Pranic		Control		Percentage	Statistics	
Traits	Mean	SD	Mean	SD	difference	t	Sig
Chlorophyll content (mg/g)	4.25	0.26	3.74	0.20	13.64	3.05	*
Plant height (cm)	53.08	6.41	43.53	3.54	21.94	4.12	*
Stem girth (cm)	1.07	0.10	0.99	0.17	08.08	1.22	NS
Root length (cm)	19.12	2.90	13.25	2.87	44.30	2.87	*
Days to first flowering	24.58	2.59	25.36	1.99	3.08	0.75	NS
Days to 50% flowering	61.75	5.50	64.5	5.00	4.26	0.73	NS
No. of days taken for first harvest	46.25	1.89	46.75	0.95	1.07	0.47	NS
No. of Fruits per plant	25.8	2.66	22.3	4.81	15.70	2.01	NS
Fruit Length (cm)	9.31	0.87	8.46	1.40	10.05	1.62	NS
Fruit Girth (cm)	1.18	0.20	1.06	0.10	11.32	1.57	NS
Fruit weight (g)	6.47	0.62	5.56	0.60	16.37	3.28	*
No. of Seeds per Fruits	64.5	10.57	51.1	4.98	26.22	3.62	*
Yield per acre (t)	14.25	0.81	12.21	0.81	16.71	3.53	*

NS, Not Significant, *Significant at 0.05 level; Values are means of ten replicates.

There was no significant variation in the days taken for flowering, 50% flowering, and first harvest between treatment and control. Yield related traits like the number of fruits per plant was 25.8 in pranic and 22.3 in control, which accounts for a 15.7% increment and the results were non-significant. Fruit length and fruit girth were also found higher in pranic treatment when compared to control, which is higher by 10 and 11% in pranic than control, but the results were non-significant. The number of seeds per fruit and fruit weight is significantly higher in pranic when compared to control. Pranic treatment had 16.37 and 26.22% improvement in number of seeds per fruit and fruit weight over the control. Yield per acre was also found significantly higher in pranic treatment by 16.7% when compared with the control plot. Pranic treatment recorded 14.3 tons yield per acre, whereas control has given 12.2 tons respectively.

RAPD marker analysis:

There were 18 scorable bands obtained with 5 polymorphic and 13 monomorphic bands (Figure 2). Percent polymorphism in primer GE2, GE3, and OPL-12 were 0, 57, and 14%. Total polymorphism was 27% with an average of 24%. Among the three primers, highest polymorphism was observed with GE3 primer and no polymorphism was seen with GE2 (Table 2). The size of the amplified product varied from 400 bp to 3000 bp. The RAPD result shows large genetic variation indicative of the evolving nature of the taxa. In a genetic diversity study in 23 chilli genotypes, RAPD marker analysis was done with 13 random primers out of that 9 were polymorphic. The percentage of polymorphic bands with different primers ranged from 14.29% to 66.67% (Bahurupe et al., 2016). In a similar study, 45 accessions of chilli were studied for variability using 16 primers using RAPD marker analysis. Three RAPD primers (decamer) efficiently amplified genomic DNA (Bhadragoudar and Patil, 2011).

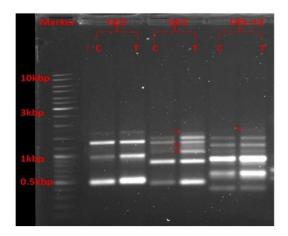


Fig. 2. RAPD banding patterns generated for Chilli using primers GE2, GE3 and OPL-12, Lane M: 10kbp ladder; C: Control, T: Treated. Red arrow indicates the presence of unique band.

Primer	Sequence of Primers (5' to 3')	Total no. of scorable bands	No. of polymorphic bands	No. of monomorphic bands	Percent Polymorphism (%)
GE2	GTTTCGCTCC	4	0	4	0
GE3	GTAGACCCGT	7	4	3	57
OPL-12	GGGCGGTACT	7	1	6	14
Total		18	5	13	27
Average		6	1.7	4.3	24

Table 2. List of primers and number of DNA polymorphism in pranic and control treatments in chilli.

DISCUSSIONS

Higher vegetative growth of the plant is an indication for the higher photosynthetic area means higher source capacity. When the plant height is more, then the chances of getting more chilli bearing branches increases which ultimately leads to higher yield (Mirza et al., 1992). A significant increase in plant height and stem girth which is an important yield contributing trait is brought about by pranic treatment. Stem girth also might have been increased by higher source capacity because of increased plant height. Plant growth and development are majorly controlled by Indole Acetic Acid (IAA) (Liscum and Reedm, 2002). The increase in plant height might be due to more cell division and formation of tissues that cause better vegetative growth of the plant that finally enhanced the plant height and stem. Increase in chlorophyll content is a direct indicator of the increase in the source capacity. The root is the source of water and minerals to the plant. An increase in yield determining traits like plant height, root length, and chlorophyll content by pranic treatment is a direct indication that it's going to bring yield changes also when applied to the crop. Most of the time flowering and fruit characteristics are varietal and genetically controlled traits and chances of getting variation by external treatment are not common. Even though flower initiation and fruit morphology are not altered by pranic treatment but the number of seeds per fruit and fruit weight is significantly increased and these are the main yield contributing traits. Seeds and the placenta holding seeds are the two important traits responsible for the pungency in chilli, which is increased by pranic treatment. Generally, stem and root are functionally interdependent and to make balance to the acquisition of both above ground (light and CO₂) and below ground (water and nutrients) resources. More root growth increases the allocation of resources to stem and leaves (Rajala and Sainio, 2001). In pranic treated plants both plant height and root length recorded at harvest were increased which might have improved in the resource acquisition from both above and below ground, further contributed for higher fruit weight, seeds, and fruit number per plant. These yield attributing traits have led to a higher yield per plant and higher yield per acre.

Pranic energy is subtle, invisible, and unquantifiable, yet has brought about significant changes in plants and the exact mechanism is unclear. So, the investigation on the exact scientific mechanism which is bringing about the changes needs to be investigated further. Pranic energy projected on plants might be received by the plant surface by receptors as a signal and the same might have been transmitted inside the plant and further physiological changes might have taken place. How the pranic energy projected affects the plant physiology and brings about morphological changes is the major thrust area. Studies in a similar area are necessary to prove the scientific reasons for the change brought about by the pranic treatment.

CONCLUSION

Chilli treated with pranic energy has exhibited enhanced plant height, stem girth, root length, and chlorophyll content. The number of seeds, fruit weight, and yield per plant were significantly increased by pranic treatment. RAPD markers recorded an average polymorphism of 24% between pranic and control. Pranic agriculture can be a supportive farming system to achieve eco-friendly natural agriculture. The use of pranic agriculture methods, in addition to regular farming practices helps in improving the yield and quality of the produce.

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تأثير الزراعة البرانية في تعزيز نمو وإنتاجية الفلفل الحار (الفليفلة الحولية) وتحليلها الوراثي

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الملخص

الزراعة برانك (PA) هي نظام زراعي تكميلي ناشئ يتم تطبيقه على البذور والتربة والشتلات لتحسين نمو النبات والمحصول من خلال تحسين جودة وكمية البرانا باستخدام تقنيات العلاج برانك. الفلفل الحار هو محصول التوابل الرئيسي في الهند. هناك طلب مستمر على الفلفل الحار في الأسواق المحلية والدولية ؛ مما يستلزم زيادة إنتاجها. أجريت دراسة ميدانية (0.2 ساعة) لفهم تأثير معالجة الزراعة البرانية على معايير نمو نبات الفلفل الحار ، والإزهار ، وخصائص الغلة والتغيرات الجزيئية على مستوى الحمض النووي عن الزراعة البرانية على معايير نمو نبات الفلفل الحار ، والإزهار ، وخصائص الغلة والتغيرات الجزيئية على مستوى الحمض النووي عن طريق تحليل واسم (RAPD). كان ارتفاع النبات أعلى معنويا (P <0.0) في معاملة البراني بنسبة 21٪ مقارنة بمعاملة المقارنة. تمت طريق تحليل واسم (RAPD). كان ارتفاع النبات أعلى معنويا (P <0.0) في معاملة البراني بنسبة 21٪ مقارنة بمعاملة المقارنة. تمت زيادة محتوى الكلوروفيل الورقي وطول الجذر بنسبة 13٪ و 4.0% على التوالي في الفلفل الحار المعالج ببرانك مقارنة بالضابطة. وزيادة محتوى الكلوروفيل الورقي وطول الجذر بنسبة 13٪ و 44% على التوالي في الفلفل الحار المعالج ببرانك مقارنة بالضابطة. وزيادة محتوى الكلوروفيل الورقي وطول الجذر بنسبة 31٪ و 44% على التوالي في الفلفل الحار المعالج ببرانك مقارنة بالضابطة. أظهرت معاملة البرانك زيادة معنوية في وزن الثمرة (6.4 جم) وعدد البذور لكل فاكهة (6.5) والمحصول للفدان (74.5 طن) أظهرت معاملة الرانك زيادة معنوية في وزن الثمرة (6.4 جم) وعدد البذور لكل فاكهة (6.5) والمحصول للفدان (74.5 طن) معان أظهرت معاملة البرانك والمحصول للفدان (74.5 طن) معان في زيادة نمو وإنتاجية الفلفل الحار المعارق والني بين ألفلاري والتعدد الأشكال 24 ٪ بين مقارنة بالكنترول (5.5 جم و 1.5 و 12.5 طن) على التوالي. سجل تحليل علامة APD متوسط وكان تعدد الأشكال 24 ٪ بين مقارنة بالكنترول (5.5 جم و 1.5 و 12.5 طن) على التوالي. سجل تحليل علامة APD متوسط وكان تعدد الأشكان والمان والني فارنة برانيك و الكنترول والخلاصة وجد أن العلاج البراني فعال في زيادة نمو وإنتاجية الفلفل الحار. ستكون الزراعة برانك نظامًا زراعيًا رانيك والكنترول والخلاصة وجد أن العلاج البراني فعال في زيادة نمو وإنتاجية الفلفل الحار. ستكون الزراعة برانك نظامًا زراعيًا والمامي والك

الكلمات المفتاحية: الكلوروفيل ,تحسين المحاصيل ,التحليل الجيني ,برانا ;شفاء برانيك، الزراعة المستدامة