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Short Communication

Phytotoxic effects of aqueous leaf extracts of *Cosmos sulphureus* Cav. on *Sorghum bicolor L*, *Vigna aconitifolia L*, *Triticum aestivum L.*

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Abstract

Weeds nearby crop fields are considered as unwanted noxious plants and exhibit many harmful effects on growth, germination and physiology of cultivated crops. *Cosmos sulphureus* Cav. is widely distributed dominant weed and is found in abundance after onset of monsoon. The present study was conducted to assess its allelopathic effects by using aqueous leaf extracts on various germination parameters of *Sorghum bicolor* L. (Moench) (jowar), *Vigna aconoitifolia L.* (moth bean) and *Triticum aestivum L.* (wheat) from Pune district. At lower concentrations (50%) stimulatory effects were observed while inhibitory effects were observed at higher concentration on germination of *Triticum aestivum* (20%), *Sorghum bicolor* (46%) *Vigna aconitifolia* (70%). Germination percentage, root length-shoot length and vigor index were significantly reduced with increasing concentrations. Root length, shoot length were at par with control at lower concentrations (50%) but as concentrations increased gradual reduction in parameters was observed in jowar and wheat, whereas moth bean exhibited moderate reduction in germination of seeds. Phytochemical analysis of aqueous crude extract showed presence of alkaloids, phenols, tannins, flavonoids, proteins, amino acids in varying amounts while saponins were absent. The variations in phytochemicals can be attributed to response of the plants to different environmental stresses as abiotic and biotic factor and mainly the geographical location. Thus, as per the results obtained *Cosmos sulphureus* exhibits allelopathic effects on germination and growth of *Sorghum bicolor*, *Triticum aestivum* and *Vigna aconitifolia* from Pune district.

Keywords: Agricultural crops, *Cosmos sulphureus*, Germination parameters, Phytochemical analysis, Phytotoxicity

'A weed is a plant growing where it is not desired or a plant out of place' (Monaco., 2002). The possibility of occurrence of any weed in non-native place could be accidental or deliberate (Bhowmik.,2014). Rice (1974) defines allelopathy as any direct or indirect effect by one plant on another plant through the production of chemical compounds that escape into the environment and influence the growth and development of neighbouring plants (Ferguson, 2003; Cheng et al., 2015). Bridges (1995) defines 'weeds are ruderals which are the plants growing in waste places, roadsides and found in highly disturbed but potentially productive environment'. As per Standard Operating Procedures (SOP) manual of Integrated Pest Management, weeds are also grouped as pests and classified according to their life cycle, mode of reproduction and habitat. Hence, prevention, control and or eradication of such noxious weeds are the major issues to be studied.

Weeds present in cultivated crop fields mainly compete with crops for sunlight, soil moisture, nutrients essential for their growth (Ghayal.,2011) through release of phytotoxic compounds which have adverse effect on growth of crops by reducing

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quality and yield of cropsbut also on biodiversity (Gupta et al., 2012., Dhole., 2013., Tomar et al., 2015). According to the research studies conducted by the authors these chemicals exhibits deleterious or harmful effects of weeds on physiological functions as cell division, cell elongation (Patil et al., 2017), inhibition of nitrogen fixation, nutrient uptake, inhibition of growth, (Joshi et al., 2015), respiration, photosynthesis, stomal opening, protein synthesis and changes in enzyme activity (Cheng et al., 2015; Baccheti et al., 2019) through release of a chemical compounds termed as allelochemicals and the phenomenon involved is termed as 'allelopathy'. Allelochemicals are present in all parts of plants but in varying concentrations and are released by different mechanism into environment through volatilisation, root exudate, leaching, plant residual matter (Siddiqui et al., 2009; Gupta et al., 2012; Sangeetha et al., 2015). The response of allelochemicals can be selective as antagonistic to one species and can be synergistic to same or another species but at lower concentrations (Tomar et al., 2015). Cheng (2015) has stated that allelopathic interactions can be one of the most significant factors in abundance and distribution of communities within and can be a successful reason for their invasiveness. Isolation and identification of such compounds is therefore today's need and integral part of research to understand the mechanism behind allelopathic interactions. Assessment of such principal compounds can be carried out by GC-MS, IR and NMR studies (Sangeetha et al., 2015).

Cosmos sulphureus Cav.an ornamental plant from Family Asteraceae with few vernacular names as Klondike, Cosmos, Orange Cosmos, Sulphur Cosmos, Yellow Cosmos (Lim., 2014). It is an annual, herbaceous, naturalized invasive weed dominant found near agricultural fields, wastelands and mainly along roadsides. Open spaces and optimum sunlight are the main factors which favour its gregarious growth attending height up to 8-10 feet. Faster growth rate and reproduction, effective seed dispersal mechanism, huge biomass and highly adaptive to different stress conditions are some of the important traits of weed for survival (Maharjan et al., 2007; Bhowmick,2014). *Cosmos sulphureus* is used as traditional medicine against *Plasmodium vivax* a malarial parasite in Brazil. It also possesses anti-oxidant, anthelminthic, anti-diabetic, hypolipidemic and anti-proliferative activity (Lim.,2014., Elodie., 2020., Raj., 2021). It has hepato-protective potential probably due to presence of quercetin, caffeic acid and chlorogenic acid (Saleem., 2017). It is a potential herbicide due to presence of sesquiterpene from leaves (Silva., 2017) and larvicidal activity (Rizki., 2013).

According to survey conducted weeds are the crop pests that cause higher reduction in yield of crop quantitively as compared to other pests. In India, loss due to weeds is 45%, insects 30%, disease 20%, and other 5%. The estimate calculated shows reduction in wheat yield is around 15-30%, 18-55% in maize, sorghum, oilseeds and pulses and 30-35% in Rice (Das, 2008). 40% of weed species are found from Family; Poaceae while rest is from Family: Asteraceae. 44 species from family: Poaceae affects crop yield mainly of barley, Wheat, sorghum, pulses, oilseeds, millets, rice whereas 32 species from Asteraceae family affects only sunflower. Such weeds have invaded crop fields either from roadside or due to crop rotation method (Jayakumar and Jagannathan, 2007).

Cosmos sulphureus possesses many medicinally important properties but, due to its presence near agricultural crop fields, studies suggests that some mechanism is involved in their invasion. Therefore, the assay was conducted to ascertain the allelopathic effects of *Cosmos sulphureus* on germination and growth of staple crops such as *Sorghum bicolor, Triticum aestivum* and *Vigna aconitifolia* by standard petri plate seed germination bioassay methods.

Collection of plant materials

The fresh leaves of dominant invasive weed *Cosmos sulphureus* were collected after the onset of monsoon during the month of September-

November at their vegetative and/ flowering stage from Mulshi Paud-Pirangut area (18^0 30' 42.30" N, 73^0 40' 49.28" E), Katraj ghats (18^0 27' 27.12" N, 73^0 52' 3.89" E) study areas of Pune district (Plate 1). The weather conditions are found suitable for growth of both crops as well as the weeds, such as rainfall, sunlight and moisture most important factors considered for growth and competition among growing plants. Soyabean, Wheat, Jowar and few leguminous crops are cultivated all over the year by rotational crop method.

The collected plant material was firstly identified and authenticated from Department of Botany, Baburaoji Gholap College, Sangvi, Pune (Auth no: Bot/ BGC-05/AUTH/2021-22). According to Aldrich (1984) leaves are the most consistent source of allelochemical than roots as latter may contain fewer and less potent toxin (Bezuidenhout., 2012). According to studies conducted leaf leachates/ extract exhibits more allelopathic effect on seed germination and other germination parameters than leachate/ extracts of root and stem (Maharjan et al,.2007).

Preparation of leaf extract

The mature fresh leaves of *Cosmos sulphureus* were collected from different parts of Pune district. They were air shade dried into homogeneous fine powder and stored until further use at room temperature. Dried plant powder was mixed with 100 ml distilled water in 500ml of beaker and kept at room temperature in dark for next 24 hours. Solution was ground by electric grinder and then filtered through muslin cloth to obtain aqueous extract stock solution of 70%. From this stock solution desired concentrations 50, 55, 60, 65, 70% (w/v) were prepared. Seeds of *Sorghum bicolor*, *Triticum aestivum* and *Vigna aconitifolia* were procured from authentic local shop. Standard petri plates (9cm diameter) were sterilized with 70% of alcohol and lined with germination paper moistened with various concentrations of leaf extract.10 seeds of test crops were placed in each petri plate for further study. Experiments were carried out in triplicates under controlled laboratory conditions during the month of June and July. Experimental control was maintained by using normal tap water. Germination papers were regularly moistened with 1 ml of desired concentrations. After 8 days germination was achieved and further parameters such as germination percentage, root length, shoot length, root-shoot ratio and vigor index were calculated. Seed germination bioassay is considered as standard and confirmatory method to define any weed as allelopathic (Rice,1984; Putnam & Tang, 1985).

Germination Parameters

Germination percentage parameter has been calculated to demonstrate number of viability of seeds for concentration of allelochemical present in nature.

Germination (%) =

$$\frac{\text{Number of seeds germinated}}{\text{Total number of seeds used for testing}} \times 100$$

Root length and Shoot length with Root: Shoot ratio are considered as authoritative parameters of developmental stages of plant indicating changes in the present environmental conditions (Sonja Gvozdenac et al., 2011) differing from plant to plant (measured in cm).

Root: Shoot ratio (cm) =

$$\frac{\text{average root length of 3 determinants}}{\text{average shoot length of same 3 determinants}}$$

Vigour Index

A vigour index test reveals seed ability to withstand different stress factors, an inversely related parameter to percentage seed germination, where seed germinates in favourable conditions.

Vigour index =

% germination x avg. shoot length of 3 determinant

Qualitative phytochemical analysis

Compounds present in plant materials are termed as "phytochemicals". Qualitative analysis of phytochemical studies is considered as one of the major analytical parts in identification of compounds such as alkaloids, tannins, cardiac glycosides, flavonoids, phenols, saponins,

phytosterols, carbohydrates and proteins. These secondary metabolites possess medicinal, insecticidal, repellent/ attractant properties. They impart colour, scent to various floral parts.

Dry plant powder weighing 5gms is soaked inDW solution and methanol separately for 48 hrs. Solution is filtered through Buchner's funnel for further evaporationof solvent and homogeneous extract was used for further phytochemical tests. Qualitative phytochemical analyses were carried out to reveal the broad groups of unknown compounds (Harborne, 1973; Kokate, 2022) (Table 4)

Statistical analysis

Calculations of germination parameters as mean average of root length and shoot length with root: shoot ratio, vigor index. All data presented are as mean values ± standard deviation along with p (0.05) value, Analysis of data results were subjected to find out overall effect of increasing concentrations on germination percetage and graphs were performed in Microsoft Excel.

Result and Discussion

Secondary metabolites are non-functional groups of compounds such as phenols, flavonoids, terpenes, tannins, alkaloids which possess specific property as plant defence, prevent from herbivory, taste and smell. All elochemicals play vital role in successful establishment of these invasive, non-native weeds as pioneer species in a highly disturbed areawith anthropogenic activities, loss of habitat, forest fires, forest degradation andin the vicinity of agricultural fields. These chemicals are released in to the environment through root exudation,leaching, leaf litter, dead matter in to the soil which affects the growth and germination of neighbouring plants. The amount of allelochemicals present in plant depends on the age, plant species, reproductive stage, location and environmental stress factors along with extraction method for preparation of leachate or extract for study. Allelochemicals have adverse effect on enzymatic activity, mobilization of nutrients, physiology of recipient plant, cell division, and finally affecting the developmental stages of seedling but at higher concentrations of leachate and plant extracts (Tomar et al., 2015., Ghayal et al., 2013). Inderjit, 2001 has mentioned in their review studies as concentration of allelochemicals, developmental stage of plant and environmental conditions affect the sensitivity of target species.

Allelopathic effects of leaf extracts of *Cosmos sulphureus* dominant invasive weed was studied on germination and growth of cultivated crops *Sorghum bicolor*, *Triticum aestivum* and *Vigna aconitifolia*. Leaf extracts had very moderate effect on *Triticum aestivum* (20%) while stimulatory or favourable for the growth of *Vigna aconitifolia* (70%) whereas, it has exhibited marginal effect on *Soghum bicolor* (46%) even at higher concentrations (70%). Inhibition or complete arrested growth of seedling may not be visible but delayed emergence of seedling growth or retarded growth (twisted form of seedlings) of both root-shoot length is observed (Kruse et al., 2000). Biochemical parameters as chlorophyll and photosynthetic pigments of *Vigna aconitifolia* were significantly decreased with increasing concentration of leaf extracts (Patil et al., 2017). These findings are similar to studies conducted earlier by (Gaikwad et al., 2020) with *Azadiricta indica* and *Eucalyptus globulus* on *Vigna* species.

Percentage seed germination

Seed germination bioassay methods are considered to be standard and confirmatory methods to define any weed 'allelopathic' according to the experimental protocols defined by Rice (1974). Germination of seed is the most susceptible parameter in allelopathic studies (Kruse.,2000; Bezuidenhout., 2012). Growth percentage, emergence of root length, shoot length, vigor index were also studied. Aqueous leaf extracts of *Cosmos sulphureus* possess moderate allelopathic effect on seed germination of all 3 studied crops with varying responses. In comparison with *Vigna aconitifolia* (70%) and *Sorghum bicolor* (46%) germination in

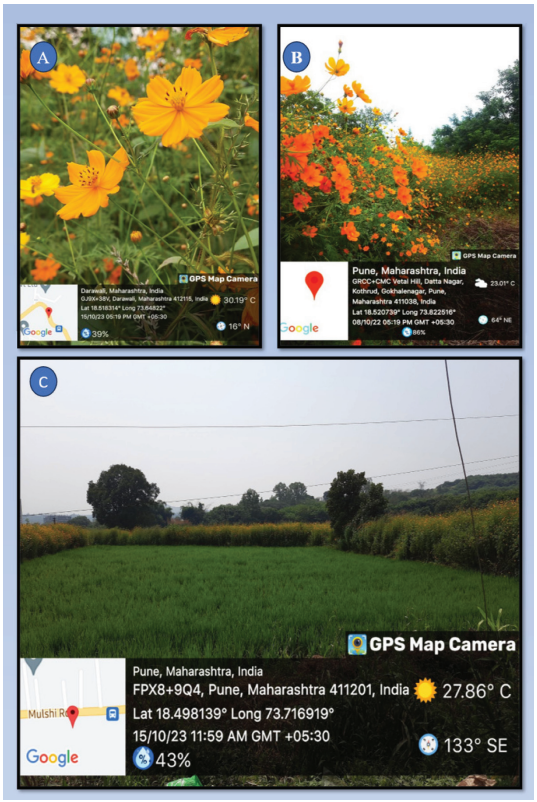

Plate 1: Invasion of *Cosmos sulphurues* near cultivated agricultural fields around Mulshi area

Triticum aestivum (20%) was affected the most at 70% concentration (Plate 1)

As per the results obtained seed germination of *Triticum aestivum* (20%) was significantly inhibited at 70% concentration, whereas studies conducted on aqueous leaf extracts of *Parthenium hysterophorus* showed maximum reduction (92.3%) at 4% concentration (Maharajan., 2007). The observations are similar with our results of seed germination results where percentage germination is directly proportional to increasing concentrations of leaf extracts. Similar results of inhibitory effects of aqueous leaf extracts of *Synendrella nodiflora* on seedling growth of wheat were recorded at 30% (Varpe., 2019). Water extracts of Yellow-Cosmos had moderate inhibitory effect on seedling growth of jowar (46%) as compared to leaf leachates of *Thlaspi arvense* L. on *Sorghum bicolor* (44.82%) at 50% concentration (Avchar., 2019). The study

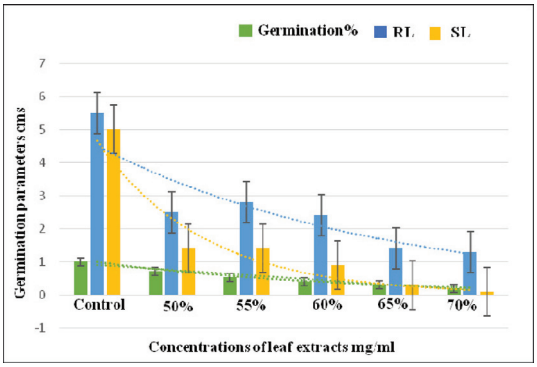

Figure 1: Effect of *Cosmos sulphureus* aqueous leaf extract on germination %, RL-root length, SL-shoot length of *Triticum aestivum*. Bar graphs showing Standard error

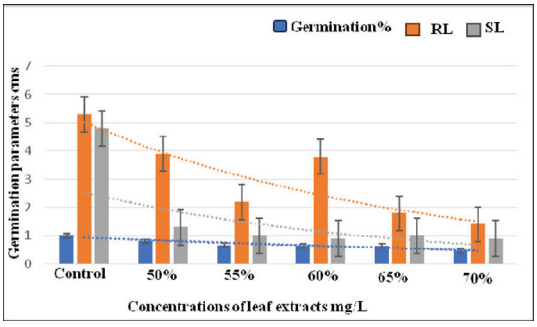

Figure 2. Effect of *Cosmos sulphureus* aqueous leaf extract on germination %, RL-root length, SL- shoot lengthof *Sorghum bicolor*. Bar graphs showing Standard error

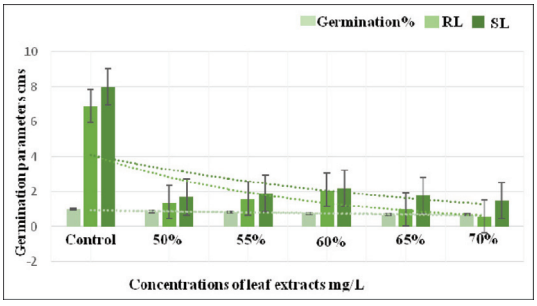

Figure 3. Effect of *Cosmos sulphureus* aqueous leaf extract on germination %, RL-root length, SL-shoot length of *Vigna aconitifolia*. Bar graphs showing Standard error

conducted revealed that leaf leachates exhibited more inhibition than root and stem extracts. *Ageratum conyzoides* restricted the germination and growth of *Sorghum bicolor* at 250, 500 and 1000

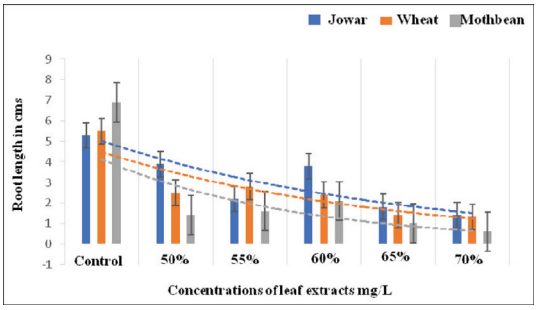

Figure 4: Comparative picture showing effect of *Cosmos sulphureus* extracts on root length of Jowar, Wheat and Mothbean

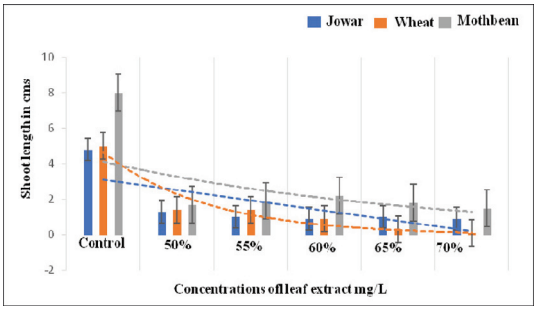

Figure 5. Comparative picture showing effect of *Cosmos sulphureus* extracts on shoot length

Table 1: The results show significant reduction in germination parameters of *Triticum aestivum* to aqueous leaf extracts of *Cosmos sulphureus*

Conc	Germination %	Root length (Mean ± SD)	Shoot length (Mean ± SD)	R:S ratio (Mean ± SD)	Vigor index (Mean ± SD)
Control	100%	5.5±0.41	5±0.38	1.1±0.08	500±37.5
50%	70%	2.5±0.16	1.4±0.09	1.7±0.11	98±6.37
55%	53%	2.8±0.15	1.4±0.08	2±0.11	74±4.0
60%	40%	2.4±0.23	0.9±0.09	2.6±0.25	36±3.42
65%	30%	1.4±0.14	0.3±0.03	4.6±0.46	9±0.9
70%	20%	1.3±0.1	0.1±0.01	13±1.0	2±0.16
p value	0.07				

Table 2: The results are indicative of effects of aqueous leaf extracts of *Cosmos sulphureus* on germination parameters of *Sorghum bicolor*

Conc	Germination %	Root length (Mean ± SD)	Shoot length (Mean ± SD)	R:S ratio (Mean ± SD)	Vigor index (Mean ± SD)
Control	100%	5.3±0.40	4.8±0.36	1.1±0.08	480±36.0
50%	80%	3.9±0.25	1.3±0.08	3±0.20	104±6.76
55%	66%	2.2±0.12	1±0.06	2.2±0.12	66±3.63
60%	63%	3.8±0.36	0.9±0.09	4.2±0.40	56.7±5.39
65%	63%	1.8±0.18	1±0.10	1.8±0.18	60±6
70%	46%	1.4±0.11	0.9±0.07	1.5±0.12	41.4±3.31
p value	0.01				

Table 3: The results have moderate allelopathic effects of aqueous leaf extracts of *Cosmos sulphureus* on germination parameters of *Vigna aconitifolia*

Conc	Germination %	Root length (Mean ± SD)	Shoot length (Mean ± SD)	R:S ratio (Mean ± SD)	Vigor index (Mean ± SD)
Control	100%	6.9±0.5	8±0.6	0.8±0.06	800±60
50%	86%	1.4±0.09	1.7±0.11	0.8±0.05	146±9.5
55%	83%	1.6±0.09	1.9±0.1	0.8±0.04	157±8.6
60%	76%	2.1±0.2	2.2±0.21	0.9±0.09	167±15.9
65%	70%	1±0.1	1.8±0.18	0.5±0.05	126±12.6
70%	70%	0.6±0.05	1.5±0.12	0.4±0.03	105±8.4
p value	0.002				

ppm concentrations (Idu 2014). Similar inhibitory effects of extracts of *Alternanthera sessilis* L. and *Cynodon dactylon* was observed at 25 and 50% concentrations, but both the weeds were stimulatory at concentrations 75 and 100% (Mali., 2014) (Table 1,2,3)

Root and shoot length

Root length, shoot length and root: shoot ratio are interrelated with percentage germination. At lowest concentration (50%) the results are at par with control but with increasing concentrations (70%) significant reduction in root length and shoot length were recorded. Significant reduction in root length was observed in *Vigna aconitifolia* (0.6 cm) than *Triticum aestivum* (1.3 cm) *Sorghum bicolor* (1.4 cm) whereas shoot length in *Triticum* (0.1 cm) was reduced at 65% concentration. mothbean (1.5 cm) showed no variation in shoot length even at higher concentrations. Varying responses were observed with all different crop seeds. (Fig 4 and 5).

In the research study conducted on of *Alternanthera sessilis* and *Ipomoea carnea* weed extracts seems to have affected the seed germination, length of plumule and radicle of *Vigna radiata* at 3 and 5% concentration (Joshi et al., 2015). Whereas, leaf extracts of *Cosmos sulphureus* had moderate effect on seedling germination and growth of *Vigna aconitifolia* with increasing concentrations (70%). The probable reason is as *V. aconitifolia* is draught resistant crop with hard seed coat and also cultivated in regions with low rainfall.

Eucalyptus globulus leaf leachate had more inhibitory effect on *Vigna aconitifolia* than *Azadiricta indica* leaf leachates at 10% concentration (Gaikwad et al, 2020). Results stated that water extract of *Argemone mexicana* had moderate effect on germination, root and shoot length of Jowar seeds. Germination in all higher concentration ranged in between 18-76 % as compared to control (89%). At maximum concentration a highest of 29% and 40% of reduction in seed germination was noticed in leaf extracts on *Sorghum bicolor* (Alagesaboopathi., 2013).

Dhole et al., (2013) reported stimulatory results of extracts of *Euphorbia hirta* followed by *Portulaca oleraceae* on *Sorghum bicolor* seed germination, growth and root-shoot length at 8% and 10%, whereas other weeds showed moderate inhibitory effects on seed germination and other parameters undertaken for study.

Root: shoot ratio

The ratio is calculated with respective values of root length to shoot length. These values are considered as suggestive parameters of developmental stages of plant indicating changes in environmental stress measured in cm. Significant changes in root: shoot ratio was observed at highest concentrations 65% and 70%.

Phytochemical analysis

Qualitative phytochemical analysis of *Cosmos sulphureus* aqueous extract showed presence of alkaloids, phenols, tannins, flavonoids, proteins, amino acids in varying amounts while saponins were absent (Table 4). These findings are corroborating with the work of Jadhav et al., (2017). Lim (2014) reported phenolic acid, gallic acid, flavonoids, protocatechuic acid, p-hydroxy benzoic acid, vanillic acid, caffeic acid, quercetin, kaempferol, myrcetin and ferulic acid as group of compounds detected from *Cosmos.*

Table 4: Qualitative phytochemicals analysis from water/ aqueous extract

Tests performed	Water/ aqueous
Alkaloid	+
Carbohydrates	+
Saponins	-
Phenols	++
Tannins	+++
Flavonoids	+++
Proteins/ amino acids	+

+++ Strongly present, ++ present, + fairly present, - not detected

Saleem et al., (2017) isolated fats-oils, carbohydrates, proteins, saponins, flavonoids, tannins, terpenoids and steroids from aqua-methanolic extracts of *C. sulphureus,* while HPLC analytical studies revealed the presence of gallic acid, quercetin, caffeic acid and chlorogenic acid. According to Fergusson (2003) the presence or absence of phytochemicals mainly depends on age of the plant, plant species, parts selected for study,

due to different geographical regions, time, and biotic-abiotic stress which are the main factors responsible for the amount and nature of allelochemicals in producer plant. Secondly, the major factor to be considered is organic solvent used for extraction method and polarity of solvent. According to author Singh (2003) phenolic compounds are water soluble which possess the allelopathic activity and they play an important role in plant-plant interactions.

GCMS and IR studies revealed presence of major constituents such as esters, ethers, anhydride and poly-alcohols. *Cosmos* showed the characteristic FTIR finger printing regions of various functional groups such as –OH, carbonyl, anhydride, ester and amides (Ghayal et al., 2018).

The study indicates that dominant invasive weed *Cosmos sulphureus* possesses moderate allelopathic effect on growth and germination of seed crops *Sorghum bicolor*, *Triticum aestivum* and *Vigna aconitifolia* from Pune district. Invasiveness of any weed is due to the presence of allelochemicals in plant which exhibits the allelopathic nature as per the environmental conditions according to the geographical location of the plant. Further research should be conducted to know the exact chemicals which possess the allelopathic property. Hence, it can be stated that such weeds can be further eradicated from agricultural crop fields and can be better source for further formulations of plant-based pesticides and insecticides.

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References

Alagesaboopathi. C. 2013. Allelopathic effect of different concentrations of water extract of *Argemone mexicana* L. on seed germination and seedling growth of *Sorghum bicolor* (L). Moench., IOSR Journal of Pharmacy and Biological Sciences., ISSN: 2278-3008,5, 52-55

Avchar, B.K. 2019. Allelopathic evaluation of *Thlapsi arvense* L. aqueous leachates on Jowar (*Sorghum bicolor*), IJRAR.,6 (2):ISSN 2348 –1269, Print ISSN 2349-5138

Bachheti, A., Sharma,A., Sharma,R, K., Bachheti,R.K., A. Husen and Pandey, D. P. 2019. Plant allelochemicals and their various applications., Co-evolution of secondary metabolites, 441-465., DOI: 10.1007/978-3-319-76887-8_14-1

Bezuidenhout, S.R.2005. Allelopathy as a possible cause for crop reduction., Agriculture and Rural development., province of KWAZULU-NATAL

Bhowmik, P.C. 2014. Invasive weeds and climate change: past, present and future., Journal of Crop and Weed,10 (2):345-349

C. Sangeetha and P. Bhaskar. 2015. Allelopathy in weed management: a critical review., African Journal of Agricultural Research, 10 (9):1004-1015., DOI: 10.1007/978-3-319-76887-8_14-1

Cheng, F and Zhiuhui, Cheng. 2015. Research progress on the use of plant allelopathy in agriculture and the physiological and ecological mechanisms of allelopathy., Frontiers in Plant Sciences, 6:1020., https://doi.org/10.3389/fpls.2015.01020

Dhole, J.A., Lone,K.D., Dhole,G.A. and Bodke,S.S. 2013. Allelopathic effect of aqueous and ethanolic extracts of some common weeds on seed health of *Triticum aestivum* L. (Wheat)., International Journal of Current Microbiology and Applied Sciences., ISSN :2319-7706 2 (6):254-260

Elodie M. M., Nyemb, N. J., N. N. Ngwasiri., A. S. Y. Fanta., F. Nveikoueing., S. F. Kouam and D. ndjonka. 2020. Invitro Antihelminthic activites of extracts and fractions of *Cosmos sulphureus* Cav against *Onchocerca ochengi*, Journal of Diseases and Medicinal Plants, 6(1): 22-30., doi: 10.11648/j.jdmp.20200601.14

Fergusson, J.J., Rathinasabapathi,B andChase,C. A. 2003. Allelopathy: How plants suppress other plants., Instt of Food and Agriculture Sciences., University of Florida

Gaikwad, S.V., Nikam,V. K andPawar,K. B. 2020. Allelopathic effects of *Azadirichta indica* and *Eucalyptus globulus* on germination of *Vigna* sp.,

Parishodh Journal., ISSN: 2347-6648., 10,1453-1457

Ghayal, N and K.N. Dhumal. 2011. Morphophysiological Investigations in Some Dominant Alien Invasive Weeds, ISBN 978-953-307-77

Ghayal, N., M. Biware and P.S. Gharpure. 2018. Phytotoxic Effects of Leaf Leachates of Invasive Weeds *Cosmos sulphureus* and *Xanthium strumarium* on Agricultural Crops., Biosciences Biotechnology Research Asia., Vol. 15(4): 821-832

Gupta, A and M. Chaabi. 2012. Effect of allelopathic leaf extract of some selected weed flora of Ajmer district on seed germination of *Triticum aestivum* L., Science Research Reporter., 2(3):311-315

Jadhav, K.M andNinge Gowda,K. N. 2017. Preliminary Phytochemical analysis and in vitro antioxidant activity of *Aracauria columinaris* bark peel and *Cosmos sulphureus* flowers., International Journal of Current Pharmaceutical Research., 9 (4): 96-99

Joshi. N., Nangia, N andJoshi,A.2015. Seed germination studies on allelopathic effects of weeds on *Vigna radiata* L., International Journal of Bioassays., 4(2):3664-3666., ISSN:2278-778X

Joshi, N and A. Joshi. 2016. Allelopathic effects of weed extracts on germination of wheat., Annals of Plant Sciences., 5(5):1330-1334

Lim, T.K. 2014.*Cosmos sulphureus*., Edible Medicinal and Non-Medicinal Plants., Vol 7, Flowers., doi: 10.1007/978-94-7395-0_19

M, Idu. 2014. Studies on allelopathic effect of aqueous extracts of *Ageratum conyzoides* Asteraceae L. on seedling growth of *Sorghum bicolor* Linn (Poaceae)., Academia journal of Agricultural Research., cabidirect.org

Kruse, M., Strandberg,M. and Strandberg,B. 2000. Ecological effects of allelopathic plants- a review., National Environmental Research Institute, NERI technical report no 15: 66

Mali, A. A and M.B. Kanade. 2014. Allelopathic effect of two common weeds on seed germination, root-shoot length, biomass and protein content of Jowar., Annals of Biological Research., 5(3):89-92

Mohammed, S., Ali, H. A., Akhtar,M.F., Saleem,U., Saleem,A. and I. Irshad. 2017. Chemical characterization and hepatoprotective potential of *Cosmos sulphureus* and *Cosmos bipinnatus* Cav., Natural Product Research, Doi: 0.1080/ 14786419.2017.143557

Monaco,T. J., Weller,S.C. and Ashton,F.M. 2002. Weed science: principles and practices., books.google.com

Patil, B.J and Khade,H. N. 2017. Allelopathic effect of *Parthenium hysterophorous* L. on photosynthetic pigments and biochemical constituents of *Vigna aconitifolia* L., International Journal of Life Sciences., 5 (4): 677-682

Raj, A. K., L.K. Bisht., D. V. Perumal., V. Candy. 2021. Pharmacological activity of *Cosmos sulphureus*, International Research Journal of Pharmaceutical and Biosciences, 5(7): ISSN 2394-5834

Rizki, I., Lalitfah dan Warlan Sugiyo. (2013) Extraction and Characterization of Bioactive Compounds in Kenikir Leaves (*Cosmos Sulphureus* Kuning) as Natural Bioinsecticide Ingredients, Indonesian Journal of Chemical Science, 2(1) ISSN 2252-6951

Silva, Bruna Pires da. 2017. Allelopathic potential of *Cosmos sulphureus* Cav., Doctoral Thesis

Singh, H. P., D.R. Batish., S.Kaur., R.K.Kohli. (2003) Phytotoxic Interference of *Ageratum conyzoides* with Wheat (*Triticum aestivum*)., Journal of Agronomy and Crop Sciences., ISSN: 0931-2250, 189:341-346

Standard Operating Procedures (SOP) for Integrated Pest Management (IPM)

Tomar, N.S., M. Sharma., R.M. Agarwal (2015) Phytochemical analysis of *Jatrophacurcas* L. during different seasons and developmental stages and seedling growth of wheat (*Triticum aestivum* L.) as affected by extracts / leachates of *Jatropha curcas* L., Physiological, Molecular and Biology of Plants., 21(1):83-92 doi:10.1007/s12298-014-0272-0

Varpe, V. D. (2019) Allelopathic effect of *Synedrella nodilfora* (L.) Gaertn on seed germination and seedling growth in Wheat (*Triticum aestivum* L.)., International Journal of Research and Analytical Research., 6(2)., e ISSN 2348 –1269, Print ISSN 2349-5138