



## Spatial distribution and sequential sampling of aphid and their natural enemies on wheat

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Although many pests attack wheat, the damage due to wheat aphids has been on rise due to climate change phenomenon. Population of aphids increases during February to March and so the ladybird beetles to devour on aphids. With the aim of facilitating conservation of ladybird beetles, predator conditioned sampling plans were worked out. Wheat aphids, *Rhopalosiphum padi* L. and *Sitobion avenae* F. were observed infesting wheat. Major predatory coccinellid beetles, *Cheilomenes sexmaculata* (Fabricius) and *Coccinella septempunctata* (Linnaeus) were also found feeding on the wheat aphids. Spatial distribution of mixed population of the aphids and coccinellid beetles was analyzed using variance-mean ratio and regression models such as Taylor's power law and Iwao's mean crowding regression. The aphids and its predatory beetles were counted at different crop age. The mean population for wheat aphid was 7.88/plant and 0.72/plant at 75 and 106 days after sowing (DAS), respectively. Variance-mean ratio indicated regular to aggregated distribution of aphids on the crop. Taylor's power law aggregation parameter ( $b = 2.62$ ) and density contagiousness co-efficient ( $\beta = 1.20$ ) of Iwao's mean crowding regression also revealed aggregated distribution of the wheat aphids. The mean population of coccinellid beetles varied from 0.72 to 1.43/plant during 68 to 100 DAS. Taylor's power law aggregation parameter ( $b = 3.64$ ) and density contagiousness co-efficient ( $\beta = 2.09$ ) of Iwao's mean crowding regression revealed aggregated distribution of the predators on the wheat crop. Sequential sampling plans were developed for wheat aphid management with and without predator's effect through Taylor's power law and Iwao's mean crowding regression. Inclusion of predator effect in the regression models increased aphid population levels which necessitates need for management measures at higher population levels. Thus, the sequential sampling plans with predator effect are useful in avoiding unnecessary pesticide application on wheat crop for the aphid management.

**Keywords:** Biological control, Coccinellid beetles, Conservation, Density contagiousness co-efficient, Iwao's mean crowding regression, Regression models, Taylor's power law, *Triticum aestivum*, Variance-mean ratio

Wheat (*Triticum aestivum* L.) is a major staple food crop, which plays a significant role in economic stability of the world<sup>1</sup>. After rice, wheat is the most important food-grain crop cultivated in India as a staple food in the Northern and North-Western parts of India<sup>2,3</sup>. India is the second largest producer of wheat in the world and accounts for 13.64 per cent of the world's total production<sup>4,5</sup>. Although many insect pests attack wheat, the damage due to aphids is increasing day by day. Aphids can cause 35 to 40 per cent direct yield losses by sucking the sap of the plants or 20 to 80 per cent indirectly through transmitting viral diseases<sup>6</sup>. Population of aphids increases during February to March and at the same time ladybird beetles like coccinellids also increase to devour on aphids. Numerous factors affect the spatial distribution of aphids such as climate, quality of host

plants, dispersal ability of aphids and their natural enemies<sup>7,8</sup> (Mann, 1995; Elliot, 2000). The term "spatial distribution is defined as the manner in which the members of a pest population are distributed in space" and it is one of the important characteristic of ecological significance of a species<sup>9</sup>. Knowledge about the temporal changes in spatial structure guides about pest arrival, its spread and helps in restricting insecticidal applications to high pest density zones<sup>10</sup>. Among insect pests, aphids are particularly prone to dispersion because they can produce numerous winged forms through parthenogenetic reproduction<sup>11,12</sup>. Integrated Pest Management (IPM) is a central theme and backbone of pest management, which uses all the available tactics to minimizing economic and environmental costs. Sampling, economic thresholds and effective tactics are the three key components of IPM methodology and are inter related. We need to have well-trained practitioners for implementing IPM components to achieve any goals

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as per national guidelines and for effective decision making<sup>13</sup>. Efficient sampling plan is indispensable for decision making in pest management. Formulation of sampling plans requires information on spatial distribution of insects and their economic injury level<sup>14,15</sup> (Stern, 1959; Binns, 1992). Sequential sampling plans needs a relatively small sample size even when insect pest's population is around or above the threshold; its saves lots of time and scouting efforts. The self-adjusting feature of a sequential plan makes it perfect and accurate for decision making.

The wheat aphid, *Rhopalosiphum padi* L. and *Sitobion avenae* F. are important upcoming pests of wheat<sup>16</sup>, especially in Northern parts of India. Nevertheless, literature available on their spatial distribution and sampling plans is scanty and needs to be studied in detail with respect to Indian conditions. Keeping the importance of aphids, predator and to have an efficient sampling plans for the management of aphids and to conserve the predatory beetles, the present studies were undertaken.

## Materials and Methods

### Field experiment

The study was conducted in experimental farm of division of entomology at ICAR-Indian Agriculture Research Institute (IARI), New Delhi. Wheat crop variety, HD 3059 was sown with 20 cm row spacing and cultivated by following recommended agronomic practices. The N, P and K was applied in the form of urea, single super phosphate (SSP) and muriate of potash, respectively. Weeds were removed manually. The crop was irrigated as per the need and no insecticides were applied on the crop.

### Spatial distribution of wheat aphids and their natural enemies

Weekly observations on population/infestation of aphids and their predators were recorded on 10 randomly selected plants in 90 plots starting from the aphid infestation till crop maturity. Mean ( $\bar{X}$ ) and variance ( $S^2$ ) of the population were estimated for each sample. Spatial distribution of wheat aphid and their coccinellid predator was analyzed using distribution parameters *viz.*, variance-mean ratio, regression model's *viz.*, Taylor's power law and Iwao's mean crowding regression.

### Variance-mean ratio

The variance ( $S^2$ ) and mean ( $\bar{X}$ ) were calculated for aphids and it's predators on all sampling dates. Value of variance mean ratio ( $\frac{S^2}{\bar{X}}$ ) = 1, <1 and >1 indicates random, regular and aggregated distribution,

respectively. The significance of departure of variance-mean ratio from unity was tested by standard normal variable (d) which is express as:

$$d = \sqrt{2 \bar{X}^2} - \sqrt{2 \bar{V}} - 1$$

Where  $\bar{X}^2 = S^2 / \bar{X}(n-1)$  and  $V = n - 1$ ; 'n' being the number of sample units. The value of  $d > 1.96$  with a negative sign indicates regular distribution. While,  $d > 1.96$  with a positive sign suggests aggregated distribution<sup>17</sup>.

### Regression models

Distribution pattern of the aphid and the predator populations were analyzed through Taylor's power law (TPL) and Iwao's mean crowding regression (IMCR) by a single regression for the entire season.

### Taylor's power law

Taylor<sup>18</sup> and Tayloret *al.*<sup>19,20</sup> showed that the variance ( $S^2$ ) of a population is proportional to a fractional power of the mean ( $\bar{X}$ ) such as:  
 $S^2 = a\bar{X}^b$

For fitting Taylor's power law to the population data of aphids and natural enemies, the variance and mean values for each observation were transformed by logarithms. The regression of  $\log S^2$  on  $\log \bar{X}$  was undertaken to estimate the parameters of Taylor's law. The value of regression coefficient (b) = 1, <1 and >1 indicated random, regular and aggregated distribution, respectively.

### Iwao's mean crowding regression

Iwao's<sup>21</sup> showed that regression of mean crowding ( $X^*$ ) on mean density ( $\bar{X}$ ) was linear and could be expressed as:

$$X^* = \alpha + \beta \bar{X}, \text{ where } X^* = \bar{X} + \left(\frac{S^2}{\bar{X}}\right) - 1.$$

The regression coefficient ' $\beta$ ' is termed as 'density contagiousness coefficient' and describe the distribution pattern of the basic components within the habitat. The value of  $\beta=1$  represent random,  $\beta < 1$  regular and  $\beta > 1$  aggregated distribution. The intercept ( $\alpha$ ) is termed as the index of basic contagion and describes the basic component of population. The value of  $\alpha < 0$  indicates that the basic component of the population is the single individual and a repulsive tendency among individuals. Whereas,  $\alpha > 0$  means that the basic component of population is a group of individuals and attractive tendency among individuals.

To estimate the parameters of Iwao's regression method,  $X^*$  was calculated for each sample and then it was regressed upon  $\bar{X}$ .

### Sequential sampling plan for wheat aphids with incorporation

**of predator effect**

Information on spatial distribution of the pest was used to formulate sequential sampling plan for wheat aphids management using.

i) Taylor's power law follows:

$$d = nm_0 \pm t \left( \sqrt{n a m_0^b} \right)$$

d = cumulative population; n = number of sampling unit;  $m_0$  = economic injury level; t = student's 't' test at 20 per cent probability level (1.28); a = sampling parameter of Taylor's power law; and b = aggregation parameter of Taylor's power law

ii) Iwao's mean crowding regression as follows:

$$d = nm_0 \pm t \left( \sqrt{n a m_0^b} \right)$$

d = cumulative population; n = number of sampling unit;  $m_0$  = economic injury level; t = student's 't' test at 20 per cent probability level (1.28);  $\alpha$  = Index of basic contagion; and  $\beta$  = Density contagious coefficient

Such that,  $d_1 = nm_0 + t \left( \sqrt{n a m_0^b} \right)$  and  $d_0 = nm_0 - t \left( \sqrt{n a m_0^b} \right)$  represented the upper and lower decision lines, respectively. Economic injury level (EIL) for wheat aphids was taken as 10 aphid/earhead<sup>22</sup>. The value of 'a' and 'b' were obtained from the analysis of spatial distribution through Taylor power law. The value of ( $\alpha$ ) and ( $\beta$ ) were obtained from the analyses of spatial distribution through Iwao's mean crowding regression.

The maximum number of samples that would be required if the cumulative number of aphids remained between the upper and lower limits, was calculated using the formula:

$$n_{max} = t^2/p^2 a m_0^b$$

Where p is 't.Sx' (t= value of normal deviate and Sx= S.E of the mean) SE of 25% of the mean was deemed as acceptable<sup>23,24</sup> and at 20% probability level, the value of 't' used was 1.28.

The above sequential sampling methodology however does not take into consideration the effect of natural enemy population on aphids, thereby suggesting need for management measures at lower population levels. Based on the predator abundance and their feeding rate, effect of predators was incorporated, so as to arrive at realistic management decision that would help to avoid unwanted pesticide application. For incorporating natural enemy effect, the equation of sequential sampling was modified to:

$$d = n (m_0 + P_e) \pm t \sqrt{[a (m_0 + P_e) b]}$$

where  $P_e$  refers to predator effect, which in turn depended upon predator density ( $P_d$ ) and its feeding rate ( $P_{fr}$ ), such that  $P_e = P_d \times P_{fr}$ . The  $P_d$  was determined through field studies while  $P_{fr}$  was used as 10.6 aphids/predator<sup>25</sup>.

**Results****Variance mean ratio**

The mean wheat aphid population varied from 0.7 to 7.9 aphids per plant during 68 to 106 days after sowing (DAS) of wheat. Variance mean ratio of the aphids during most observations, except at 75 and 82 DAS was found to be <1 indicating regular distribution. The population variance of aphids increased with an increase in its density. The highest variance (11.5) was noticed at the highest aphid density (7.9) (Table 1). The mean coccinellid predator population varied from 0.71 to 1.43 coccinellid per plant during 68 to 106 DAS of wheat crop. The population variance of coccinellids increased with increase in their density. The highest variance (1.22) was observed at the highest coccinellids density (1.43) (Table 2).

**Regression model:**

The aggregation parameter (b=2.62) of Taylor's power law significantly >1 during the crop season revealed the aggregated nature of the aphid population (Table 3).

The regression equation of Taylor's power law for wheat aphid population has been found as:

$$\log S^2 = 2.62 \log X - 1.01 \quad (R^2 = 0.86)$$

The Taylor's power law equation for variance and

Table 1 — Variance - mean ratio and standard normal variable (d) for wheat aphid

Crop age (DAS)	X	S <sup>2</sup>	S <sup>2</sup> /X	Standard normal variable (d)
68	2.482	2.1284	0.8575	- 0.9866
75	7.880	11.464	1.4548	2.7505
82	5.237	7.6623	1.4631	2.7963
91	1.329	0.3679	0.2769	- 6.32104
100	1.060	0.3203	0.3021	- 6.0086
106	0.717	0.0084	0.0117	- 11.8985

Table 2 — Variance - mean ratio and standard normal variable (d) for coccinellid on wheat

Crop age (DAS*)	X	S <sup>2</sup>	S <sup>2</sup> /X	Standard normal variable (d)
68	0.713	0.001	0.0041	- 12.4874
75	0.769	0.031	0.0410	- 10.6401
82	0.767	0.034	0.0447	- 10.5209
91	0.805	0.041	0.0510	- 10.3287
100	1.428	1.228	0.8593	- 0.9742
106	1.060	0.0001	0.0001	- 13.2082

Table 3 — Parameter of TPL for aphid and coccinellid population on wheat

Pest/ Predator	No. of samples (n)	Index of basic Contagion ( $\alpha$ )	't' value	'p' value	Density contagiousness coefficient ( $\beta$ )	't' value	'p' value	Coefficient of determination ( $R^2$ )
Aphids	6	0.123	-5.429001	0.005584	1.204	29.20363	8.18E-06	0.99
Predator	6	0.014	-6.59412	0.00274	2.087	7.170836	0.002002	0.93

Table 4 — Parameter of IMCR for aphid and coccinellid population on wheat

Pest/ Predator	No. of samples (n)	Sampling parameter (a)	't' value	'p' value	Aggregation parameter (b)	't' value	'p' value	Coefficient of determination ( $R^2$ )
Aphids	6	0.0993	-3.8351	0.0185	2.6227	5.03764	0.00729	0.8638
Predator	6	0.0254	-2.5606	0.0626	3.6361	0.68023	0.53369	0.1037

mean relationship for wheat aphid was computed as,  $S^2 = 0.0993 X^{2.62}$ . The aggregation parameter, ( $b = 2.62$ ) suggested aggregated distribution of the pest in the field. The value for coefficient of determination ( $R^2$ ) revealed that the mean aphid population accounted for 86 per cent variation in population variance. The regression equation for Iwao's mean crowding regression for wheat aphid population was calculated as:

$$X^* = 1.21x - 0.91 \quad (R^2 = 0.995)$$

The density contagiousness coefficient ( $\beta = 1.204$ ), also indicated aggregated distribution of aphids during the crop season. The index of basic contagion value ( $\alpha = 0.123$ ) (Tab. 4) revealed that the basic component of the population was a group of aphids having attractive tendency among them. Taylor's power law equation for coccinellids was found to be:  $\log S^2 = 3.64 \log X - 1.59$  ( $R^2 = 0.104$ )

The Taylor's power law thus did not explain relationship for coccinellids satisfactory as indicated by low  $R^2$ . The equation for Iwao's mean crowding regression for coccinellids was found as:

$$X^* = 2.096x - 1.84 \quad (R^2 = 0.93)$$

The value of density contagiousness coefficient ( $\beta = 2.096$ ), indicated, aggregated distribution of coccinellid during the all crop stages. The index of basic contagion value ( $\alpha = 0.015$ ) evinced the basic component of the population was group of coccinellids that showed an attractive tendency among them (Table 4).

#### Taylor's power law based sequential sampling:

##### Without predator effect

Sequential sampling decision line with aggregation parameter ( $b = 2.62$ ) and sampling parameter ( $a = 0.099$ ) of Taylor's power law, economic injury level as 10 aphids/head and tolerable error as 20 per cent ( $t = 1.28$ ) was computed as:  $d = 10 n \pm 7.84\sqrt{n}$ . The value of lower and upper decision lines after observing three sample unit (for example), as 17 and 44 aphids/plant, suggesting no action and action,

Table 5 — Sequential sampling plan through TPL for treatment decision against wheat aphid without and with predator's effect

No. of Sample (n)	Without predator's effect.		With predator's effect.	
	Lower decision line $d_0 = 10 n -$ $7.84 \sqrt{n}$	Upper decision line $d_1 = 10 n +$ $7.84 \sqrt{n}$	Lower decision line $d_0 = 19.54 n -$ $18.854 \sqrt{n}$	Upper decision line $d_1 = 19.54 n +$ $18.84 \sqrt{n}$
1	2	18	1	38
2	9	31	12	66
3	17	44	26	91
4	25	55	40	116
5	31	69	56	139
6	41	79	71	163
7	49	90	87	187
8	58	102	103	210
9	66	113	119	232

respectively (Table 5 Fig. 1A). However, the population between 17-44 would require more sampling until decision to take management action is arrived at. However, in case of indecisiveness, the maximum sample size was determined to be seven sampling units and if decision is not reached even after 7 sampling units, then sampling would be suspended and resumed after 4-5 days interval.

##### With predator effect

Likewise, decision lines of sequential plan for aphids with inclusion of predator effect was computed as:  $d = 19.54 n \pm 18.854 \sqrt{n}$ . The value of lower and upper decision lines after observing three sample unit was 26 and 91 aphids/plant, indicating no action and action, respectively (Table 5 and Fig. 1B). The decision line with predator effect thus suggested need for action at higher aphid population compared to without predator effect.

#### Iwao's mean crowding regression based plan

##### Without predator effect

Sequential sampling decision line with density contagiousness coefficient ( $\beta = 1.204$ ) and index of basic contagion ( $\alpha = 0.123$ ) of Iwao's mean crowding regression, economic injury level as 10 aphids and tolerable error in decision as 20 per cent ( $t = 1.28$ )

Table 6 — Sequential sampling plan through IMCR for treatment decision against wheat aphid without predator’s effect

No. of Sample (n)	Without predator’s effect.		With predator’s effect.	
	Lower decision line	Upper decision line	Lower decision line	Upper decision line
	$d_0 = 10 n - 5.4912 \sqrt{n}$	$d_1 = 10 n + 5.4912 \sqrt{n}$	$d_0 = 19.54 n - 18.854 \sqrt{n}$	$d_1 = 19.54 n + 18.84 \sqrt{n}$
1	5	15	7	32
2	12	28	21	56
3	20	40	36	79
4	29	51	52	101
5	38	62	67	124
6	47	73	85	146
7	55	85	102	167
8	64	96	119	189
9	74	106	136	210

were calculated as:  $d = 19.24 n \pm 5.49 \sqrt{n}$ . The value of lower ( $d_0$ ) and upper decision ( $d_1$ ) lines after observing three sample unit (for example) was 20 aphids and 40 aphids/plant, indicating no action and action, respectively (Table 6 and Fig. 2A). However, the population between 20-40 would require more sampling until decision about management is taken.

*With predator effect*

Similarly, the decision line of sequential plan for aphids with the inclusion of predator effect was computed as:  $d = 19.54 n \pm 12.34 \sqrt{n}$ . The value of lower and upper decision lines after observing three sample unit (for example) was 36 and 79 aphids/plant, indicating no action and action respectively (Table 6 and Fig. 2B). The decision line with predator effect thus suggested need for action at higher aphid population.

**Discussion**

Wheat is attacked by many insect pests, but, the damage due to aphids has been increasing in recent years. The wheat aphids, *R. padi* and *S. avenae* occupy different ecological niches on the same host plant. Normally, *R. padi* arrives first, prefers the stem and lower leaves and attains peak before *S. avenae* arrives. However, *S. avenae* is found mostly on the ears and upper leaves of the plant<sup>26</sup>. An environmental variation in both space and time is a major factor which influences the dynamics and distribution of many animal species<sup>27,28</sup>. Efficient sampling plan is required for decision making in insect pest management. Sampling plan needs information on spatial distribution of insect pests and their economic injury level for its formulation<sup>29</sup>.

Spatial distribution of the aphids and coccinellid

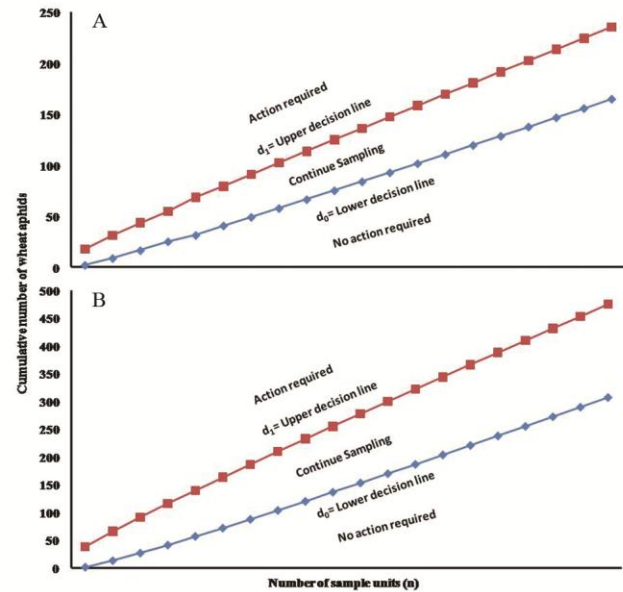


Fig. 1 — Sequential sampling plan through TPL for treatment decision against wheat aphid (A) without predator’s effect; (B) with predator’s effect.

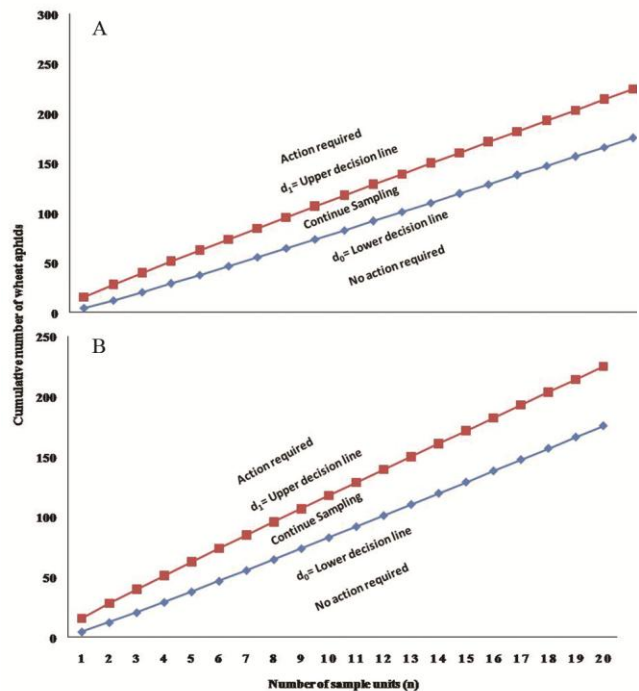


Fig. 2 — Sequential sampling plan through IMCR for treatment decision against wheat aphid (C) without predator’s effect; (D) with predator’s effect.

predators was analyzed through variance-mean ratio and regression models viz., Taylor’s power law and Iwao’s mean crowding regression. The aphid species viz., *R. padi* and *S. avenae* were recorded from wheat, which were also reported earlier from Northern plains

of India on wheat<sup>30</sup>. Incidence of wheat aphids began at 68 DAS and peaked at 75 DAS with maximum variance. Aphid population in the field was regularly distributed at the beginning and end of the crop season. However, during 75 and 82 DAS, the aphids were distributed in an aggregated fashion as revealed by variance-mean ratio and standard normal variable (d). Our results are in concurrence with Borges<sup>31</sup> and Tomanovic<sup>32</sup> who have also observed random to aggregated distribution of cereal aphids *viz.*, *R. padi* and *S. avenae*. The Taylor's power law ( $b = 2.62$ ) and Iwao's mean crowding regression ( $\beta = 1.204$ ) also revealed aggregation as dominant pattern of aphids distribution during the wheat crop stages in the field. Aggregation of aphids was also reported earlier by Godfrey and Chaney<sup>33</sup>, Tomanovic *et al.*<sup>34</sup> using regression models on celery plants and cereals. The predatory coccinellid beetle species *viz.*, six spotted ladybird beetle, *Cheilomenes sexmaculata* (Fab.) and seven spotted ladybird beetle, *Coccinella septempunctata* (L.) were observed feeding on the wheat aphids. Coccinellids as an efficient predator of cereal and other aphids have also been reported earlier<sup>35,36</sup>.

Studies on spatial distribution of insect pests and their predators have been undertaken earlier too<sup>8</sup>. During most of the crop stages and at different aphid densities, the coccinellid predators were distributed regularly ( $S^2/\bar{X} \leq 1$ ). It might be due to existing cannibalism and intraguild predation among the predators; such avoidance behaviour leads to spatial segregation between predator species for reducing interaction to facilitate co-occurrence for future survival<sup>37</sup>. The values of standard normal variable (d) greater than  $-1.96$  for coccinellid predators during most of the crop stages indicated regular distribution of the predators. Taylor's power law regression ( $R^2 = 0.10$ ) did not provide good fit to coccinellids population data whereas, Iwao's mean crowding regression ( $R^2 = 0.93$ ) did it. The index of basic contagion value ( $\alpha = -1.59$ ) indicated that there was repulsive tendency among the predators due to cannibalism and intraguild predation. These tendencies among the predators are governed by the temporal and spatial distributions of aphids and other sucking pests<sup>38</sup>.

Sequential sampling helps taking quick decisions in insect pest management. It takes into account spatial distribution and economic injury level of insect pest on the crop<sup>13</sup>. Sequential sampling classifies the insect

pest population into those warranting control actions and those not warranting control actions. Development of sequential sampling plans with the incorporation of natural enemy effect has been ignored for undertaking management decision for so long. Of late, lots of works have been conducted in devising sequential sampling plans for wheat aphids but without predator effect. However, an attempt is made to include coccinellid predators in decision making in present study. Coccinellid beetles are important predator in agri-horticultural crops and have been used as a bioagents against a number of sucking pests because of their voracious feeding habits<sup>39</sup>. Natural enemy population normally builds with the population of its pests and there exists a correlation and general equilibrium between insect population and its natural enemies<sup>40</sup>. Sequential sampling plan with inclusion of coccinellid population with TPL as well as IMCR suggested need for control action at higher aphid population than when coccinellid population was not considered, leading to avoiding unwarranted pesticide application, coccinellid conservation, environmental safety and attractive benefit cost ratio.

Comparison of sequential plans with TPL and IMCR revealed that both suggest action after three sampling unit at cumulative aphid population of 91 and 79, respectively. Sequential plan with TPL was thus observed to be slight better in delaying pesticide application compared to IMCR. There is always a debate on which model fits better, and it depends on which data sets are exposed to and also different assumptions and approaches behind these two models. TPL is essentially adopted based on its fit to a wide range of field data compared to Iwao's patchiness regression<sup>41</sup>. Taylor expressed that the parameters of his model would be unique to each species and constant over all ranges<sup>18</sup>. Predator conditioned sampling plan would thus help in avoiding excess pesticide application, thereby facilitating conservation and utilization of natural enemies and ensuring better benefit cost to growers<sup>42</sup>.

## Conclusion

Information on dispersion pattern of the aphid and coccinellids predators would be helpful in better utilization of coccinellids for the biological suppression of wheat aphids. Also, the information about the physical location of the aphid gives clue about its arrival, spread and also in concentrating

pesticidal applications on plants having high aphid density. Devising sequential sampling plan saves time, labour and plan with predator effect would also save the cost on pesticide use by limiting the treatment of pesticides. Sequential sampling plan would thus help the grower's in conserving the populations of predatory coccinellid beetles.

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### Conflict of interest

The authors declare no conflicts of interest.

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