EVALUATION OF RESIDUAL ACTIVITY OF VARYING CONCENTRATION OF INDAZIFLAM AS IT PERSISTS IN THE SOIL.

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ABSTRACT
The study determined the residual effect of Indaziflam herbicide on some common arable crops, intercropped within the palm inter-rows,as the herbicide persist with a view to providing information on affecting proper weed management strategies in oil palm cropping systems.The experiment was conducted in a greenhouse of main station of Nigerian Institute for Oil Palm Research (NIFOR) near Benin City($6^0$3'N and $5^0$37'E) in 2016 and 2017 cropping seasons.The experimental design was complete randomized design in three replicates per species for each dose.The Bioassays method was used to evaluate the residue persistence for some selected oil palm inter crops. It was observed that indaziflam negatively affected the growth of maize, melon vein length, cucumber vein length and tomato as the concentration of indaziflam was increased. The predicted dry weight increased as the storage time was increased. In conclusion, the period of four weeks for indaziflam persistence was not sufficient for maximum growth performance of maize, melon, cucumber and tomatoes-oil palm inter crops.
Keywords-Concentration, Indaziflam, residue activity, persistence, soil.

INTRODUCTION

Oil palm (*Elaeis guineensis* Jacq.) is native to tropical Africa and it is an economic crop in Central, South America and South East Asia. In West Africa oil palm is cultivated along the coast of Sierra Leone, Liberia, Ivory Coast, Ghana, Togo, Benin, Cameroons and Nigeria (Ekhator *et al.*, 2018; Corley & Tinker,2003). In these countries, oil palm is cultivated to meet the local and growing industrial demand for palm oil and palm kernel. Oil palm is the highest oil- producing crop in the tropics with potential yield capacity of more than 10 tons of oil per hectare (ha^{-1}). However, current yields in the world are well below 10 tons' ha^{-1} and are actually about 4-6 tones ha^{-1} for the best managed commercial estate and 3-4 tons' ha^{-1} for the managed smallholders' farms (Murphy, 2014) However, weed management is a major agronomic and intensive problem in the cultivation of oil palm across West and Central Africa. Weed competition is a serious constraint to oil palm production. The cost, time, and frequency of weed control are contingent upon the types of weed present. Weed species compositions across the countries in West Africa are closely related (Akobundu *et al.,* 2016). The high infestation and frequent regrowth of weeds increase labour costs and inputs such as herbicides in managing plantations for cost efficiency and profitability. The use of combined herbicides mixtures such Indaziflam and glyphosate is known among small holders of oil palm in the control of weed in their plantation. Studies have shown the persistence of these herbicides combination in the soil (Peres-Oliveira *et al.,* 2017). The length of time some of these herbicides remain active in the soil could be long and their after effects may prove injurious to succeeding crops or plantings. Herbicides persistence is an important aspect to be considered in oil palm production because in oil palm cultivation, food crops (arable) are sometimes incorporated as intercrop and residues of applied herbicides can potentially injure sensitive crops grown as intercrop. It is difficult to predict the amount of herbicides in the soil and their residual effects on arable crops grown within the palm rows at the early stage of field planting. Herbicides residues cause great variability in plant growth and quality and in severe cases, can result in complete crop loss and high economic loss to oil palm growers. Bioassay provides practical and acceptable information on the detection of low level of herbicides in soil (Pestemer *et al*., 1980).

MATERIALS AND METHOD

Evaluation of the residual activity and persistence of the indaziflam in the soil of different concentrations in the soil

The experiment was conducted in a greenhouse of mainstation of Nigerian Institute for Oil Palm Research (NIFOR) near Benin City($6^0$3'N and $5^0$37'E) in 2016 and 2017 cropping seasons. NIFOR Benin city research station is characterized by a long rainy season and a short dry season with a bimodal rainfall distribution. Temperature ranges between

21.3℃ (minimum) and 34.7℃(maximum) with an altitude of 149 m above sea level. Soil used was analyzed with standard procedures.The experimental design was the complete randomized design in three replicates per species for each dose.

Development of bioassay techniques for evaluating the residual activity of indaziflam

The procedures for establishing bioassay trial was the procedures developed by Horowitz (1976) and Eliason *et al*. (2004) which were adopted for the conduct of greenhouse bioassay.Soils were collected from NIFOR herbicide free plots (Field 30)that were used. Sampling procedure involved the use of 7.5 cm soil auger to collect soils at 0-15cm depth. Sampling was done randomly in diagonal transects measuring 9m. Eight soil core samples were taken along the diagonal transects. A total of 7.2 kg of sampled soils were divided into 2.4 kg per three replicates of each test crop. The soil samples were air-dried at room temperature for 48 hours after the soil was then screened through a 4mm sieve to remove stones and large pieces of organic matter. Water was continuously added to completely wet the soil without leaving standing water in the bottom of the cup. The soil field capacity was determined by using methods similar to Eliason *et al*. (2004). Then the saturated soil was weighed and soil field capacity was calculated as the ratio of the

$$\frac{\text{Weighed: weight of saturated soil (g)} - \text{weight of dry soil(g)}}{\text{Weight of dry soil (g)}} \times 100\ \%$$

The soil field capacity was calculated to ensure that each experimental unit for the greenhouse bioassays was maintained at 80% field capacity. Stock solutions for the treated-soil bioassay were prepared by developing a desired concentration in part per million then, 1ppm = ml/L (Streibig, *et al.,* 1993). To calculate for different indaziflam concentration the herbicide 1ml of indaziflam herbicide dissolved in 1L = 1000 ppm. But, 1ml of the indaziflam herbicide (being a liquid with specific gravity = 1.10) =M/D =1/1.10 =0.9mls.Therefore, 0.9mls of herbicide in 1liter of water = 1000 ppm. However , application of the serial dilution of the above proved lethal to the plant, hence 0.2mls of the indaziflam herbicide was used and dissolved in 1L of distilled water which gave a concentration of 220ppm, if 0.2mls indaziflam herbicide made up to 1000mls water, with stock solution of 200ppm which formed a new stock solution of 200ppm serial dilutions were made using the formular:

$$C_1V_1 = C_2V_2$$

$= V_2 = \frac{C_1V_1}{C_2}$ (www.Kent. a.c uk/student-learning-advisory -service)

Where C_1 implies initial concentration.

V_1 Implies the initial volume,
C_2 Was final or desired concentration
V_2 Was the desired volume which obtained by the formula

However, stock implies 1ml/L, then the stock was left at room temperature for less than 24 hours before being applied to the soil. In order to spike or treat the untreated soil collected from the field for indaziflam concentrations, the different concentration at (0, 0.01 ml, 0.02 ml, 0.03 ml, 0.04 ml, 0.05 ml, 0.06 ml, 0.07 ml, 0.08 ml, 0.09 ml, 0.10 ml with their equivalent in active ingredient per hectare (0.0045, 0.0090, 0.0135, 0.0180, 0.0290, 0.0270, 0.0315, 0.0360, 0.0360, 0.0405 and 0.045 kg a.i ha-^{1}) indaziflam were transferred from the stock solution and added to 1000m/l the flasks. Distilled water was added to the indaziflam solution to reach a total volume of 1000 mls, 2.4kg of soil from the untreated field plots was laid out evenly on a tray lined for each indaziflam dose. The soil was spiked by slowly adding each solution using a Gustafson® Batch Lab Treater. The spiked soils for different concentrations were left sealed in the bags for 14-16 hours at room temperature. Soil was again mixed thoroughly by hand. The herbicide-treated soils were divided into 2.4kg portion which was dispensed in 7cm deep bioassay cups perforated at the bottom. Each cup was sub-irrigated to capacity and allowed to consolidate before sowing ten seeds of test crops at depth of 1cm and covered lightly.

Data Collection

Plant height, was collected at the expiration of 21days after emergence, thenthe whole plant of the experimental test crops (maize, cucumber, melon and tomatoes) were harvested by cutting the plant at the base and dried to a constant temperature for the dry weight determination.The mean percentages loss in growth parameters of all the test crop fresh / young shoots in each treatment across replicates were calculated as follows:

$$\%\ \text{Growth loss} = 100 - \left(\frac{A}{B}\ x\ 100\right)$$

Where 100 = percentage of the maximum value of growth parameter expected, A = value of growth parameter in un-treated herbicides (test plants) while B = value of growth parameters in the treated herbicides plots. The mean data percentage loss in growth parameters from each of the treated plants across the three replicates for each years of study was subjected to a non-linear regression model as described by (Streibig, et al., 1993) (see equation below) to determine the functional relationship between the dependent variablewhich was the x= the herbicide dose and independent variables was y = which the time the herbicide persisted either in the field or in the soils. The non – linear regression was used to fit appropriate curves to predict the loss of test crop at every point in the curves and
The logistic equation in explicit was

The equation is:

y = a + c / (1 + exp (-b* (x – m)))

y = total test crop vigor, a = seedling (test crop) vigor from herbicides treated soil, b = change in (test crop) vigor due to change in time of storing treated soil or time herbicides persisted before sampling.c = the eventual test crop vigor from treated soils. m = theoretical value predicated by the model or the speed at which treated herbicide residues soils affects test crop vigor, x = time of storing or sampling .

Statistical Analysis

Analysis of Variance (ANOVA) was performed on the heightand dry weight using the GenStat Release 12.1(2012) edition and differences between two treatment means were compared using standard error of themeans difference at 5% level of probability.

RESULTS:

Residual effects of indaziflam concentration weeks after storage treatment on height of tomato, cucumber vein length, melon vein length and maize

Indaziflam concentrations in soil medium at different storage time significantly affected the height of tomato seedlings 21 days after planting (DAP) in 2016 and 2017(Table 1). Tomato height decreased considerably as the concentration of indaziflam increases from 0.0 - 0.045kg a. i ha^{-1} irrespective of the storage time. The height of tomato in the control plot was significantly different from all other treatments in both years(Table 1).Indaziflam concentrations in soil medium at different storage time significantly reduce vine length of cucumber seedlings 21 days after planting (DAP) in 2016 and 2017 (Table 3). Cucumber vine length was negatively affected as the concentration of indaziflam increases from 0.0 kg a.i $ha^{-1}$to 0.045kg a.i ha^{-1}irrespective of the storage time . Seedlings length of melon decreased as the concertation of indaziflam increases from 0.0 to 0.045kg a.i ha^{-1} irrespective of the storage time. Indaziflam at 0.045 kg a.i ha^{-1} at 0 WST caused stunting of melon length by 95% in both years(Table4). Indaziflam concentration significantly affected length of maize seeding irrespective of the storage time(Table 5). Height of maize seedling was 14.383 cm and 14.897 cm at 0.0 kg a.i kg $ha^{-1}$of indaziflam at 0WST in 2016 and 2017. Similarly, height of maize seedling was 38.33 cm and 38.12 cm at 0.0 kg a.i ha^{-1} soil at 4WST. Furthermore, stunted maize height of 10.49 cm and 10.28 cm was observed at indaziflam concentration of 0.045 kg a.i ha^{-1} of WST. The concentration of indaziflam at 0.0 kg a. i ha^{-1}soil irrespective of the storage time was significantly higher than other treatments(Table 5).

Predicted Loss in Maize cucumber, melon and tomato Dry Weight Due to Indaziflam Concentration in Soil Weeks after Storage Treatment

Maize,cucumber, melon and tomato whole dry weight loss at different concentration of indaziflam fitted inthe logistic regression model with coefficient of multipledetermination (R^2) value of 98% (Fig.1,2,3,4.).Loss in dry weight of maize, cucumber, melon and tomato increased as indaziflam concentration in the soil was increased.Indaziflam concentration in soil at varying weeks of storage treatment significantly affected dry weight of maize (Fig. 1). At 0WST, increase in the concentration of indaziflam from 0.009 kg a. i ha^{-1} indaziflam to 0. 045kga.i ha^{-1} resulted in maize dry weight loss of between 11.7% to 26.7 % respectively(Fig.1,2,3,4). As atthe time of storage increased losses in dry weight of maize, cucumber, melon and tomato decreased (Fig.1,2,3,4). Cucumber dry weight loss varied with indaziflam concentration and storage time significantly. Cucumber dry weight loss was 43% at indaziflam concentration of 0.0045 kg a.i ha^{-1} at 0WST while at 4 WST loss in dry weight of cucumber was minimal (Fig 2). Loss in dry weight of melon and tomato was significantly affectedby indaziflam concentration and storage time(Fig.3&4).

DISCUSSION:

Residual effect of indaziflam concentration and storage time on tomato, cucumber, melon and maize growth

The observed decrease in tomato height, cucumber vine length, melon vine length and maize height as the application rate of indaziflam increased in the soil medium of the test crops is an indicative of the phyto-intoxication of indaziflam to the test crops when usually sown in succession or the carryover effect of indaziflam. Carryover effect of diuron and fomesafen has previously been shown to affect bean, maize and soybean sown in succession to their application in cotton (Gheno *et al*., 2016). Similarly, fluometuron [*N, N*-Dimethyl-*N'*-[3-(trifluoromethyl)-phenyl]] urea was found to injured soybeans if a waiting period of 4 to 6 weeks after an application is not observed (Corbin *et al*., 1994). The negative effect of indaziflam on tomato, cucumber, melon and maize up to four weeks after sowing the test crops is an indication that this herbicide could persist in the soil with long bioactivity that may cause subsequent damage to other sensitive crops for a longer period. The persistent nature of indaziflam could be attributed to its limited mobility in soil medium as reported by Jhala and Sigh (2012). Several herbicides used in oil palm and most especially soil - applied herbicide such as metsulfuron and diuron have been shown previously to persist in the soil for a longer period after application (Ekhator *et al*., 2018).

Predicted loss in maize, cucumber, melon and tomato due to indaziflam application rate andvaried storage time in soil

The predicted increase in the negative effect of indaziflam on the test crops as the level of indaziflam concentration increases in soil medium is an indication that dosage is an important factor in indaziflam phyto intoxication. Similarly, 2, 4-D dosage has been shown to significantly affect soy bean growth (Peres-Oliveira *et al.,* 2017). The predicted decrease in dry weight loss of the test crops as the storage time of sampled indaziflam treated soil was increased is an indication of degradation and dissipation of indaziflam in soil. Herbicide degradation and dissipation in soil have been widely reported (Williams, 2001; Jed, 2006). The loss sustained at 4 weeks of soil storage after herbicide application is evident that indaziflam will have a carryover effect in successive crops. A previous study has buttressed this fact that the amount of herbicide applied in soil and the susceptibility to crops determine if injury to a rotational crop will occur and if most crops will not be susceptible at a minimum tolerable application rate (Williams, 2001). Similarly, the application of imazequin to soybean at 1.5 to 2.0 times the recommended rate was reported to have increased its chance of carryover injury.

Table2.Residual effect of different concentrations of indaziflam tomato (cm) seedlings
$0_{4}^{\dagger}$Weeks after storage of soil treated with indaziflam,

Dose rate (kg a.i /ha-^{1})	2016					2017				
	0	1	2	3	4†	0	1	2	3	4
0.000	4.000	4.300	4.044	4.256	4.400	4.013	3.470	4.013	4.049	4.400
0.004	2.670	2.663	3.560	3.599	3.547	2.650	2.678	3.286	3.600	3.634
0.009	2.513	2.617	2.904	3.511	3.340	2.340	2.730	2.970	2.949	3.340
0.013	2.403	2.213	3.340	3.340	3.340	2.120	2.138	3.007	3.650	4.167
0.018	1.360	1.510	3.014	2.674	3.107	2.023	2.088	3.660	3.566	4.478
0.020	1.394	1.640	3.004	2.699	3.270	1.502	1.944	3.173	3.300	3.300
0.027	0.668	0.763	2.080	2.154	2.340	1.673	1.681	2.089	2.956	2.318
0.031	0.661	0.853	2.130	2.053	2.322	0.633	0.837	2.107	2.140	2.321
0.036	0.510	2.033	2.106	2.122	2.289	0.510	0.844	2.116	2.120	2.312
0.040	0.144	0.213	2.100	2.344	2.222	0.144	0.244	2.113	2.130	2.390
0.045	0.144	0.210	0.334	1.091	1.318	0.144	0.244	0.300	1,022	1.340
‡SED (interaction)			0.1297					0.1858		

‡ Standard error comparing dose rates and soil storage time after indaziflam applicati

Table 3. The residual effect of different concentrations of indaziflam on vine length of cucumber(cm) seedlings at 0 to 4 weeks after treatment in 2016 and 2017

Dose rate (kga.i/ha-^{1})	**2016**					**2017**				
	0	**1**	**2**	**3**	**4**†	**0**	**1**	**2**	**3**	**4**
0.000	3.3233	4.7013	6.3623	15.1100	18.1233	3.244	4.706	6.369	14.950	18.047
0.004	3.2157	4.3183	6.6400	9.9623	10.2890	2.836	4.307	5.387	10.350	11.330
0.009	3.0800	4.3097	6.2400	9.6040	10.0120	2.536	4.217	5.060	9.914	10.012
0.013	3.0800	3.4433	5.2633	9.8623	9.9027	2.339	3.618	5.114	9.882	10.008
0.018	3.0013	3.6277	5.2343	9.0227	9.0387	2.253	3.531	5.158	6.335	9.950
0.020	2.0157	2.5093	5.1667	6.3400	9.0023	2.004	2.503	5.104	5.910	9.598
0.027	0.4767	1.5073	3.7073	5.8210	9.0021	0.327	1.507	3.013	5.322	9.367
0.031	0.3047	1.3243	3.6773	5.3287	8.5673	0.200	1.223	2.494	2.301	7.283
0.036	0.2203	1.0073	2.2343	2.0183	3.6770	0.734	1.134	2.222	2.303	3.210
0.040	0.1610	0.6800	2.0313	2.0183	3.6780	0.490	0.683	2.001	2.024	3.623
0.045	0.1117	0.6757	2.0313	2.0257	3.6723	0.129	0.652	1.964	2.001	3.648
‡SED(interaction)			0.5481					0.3750		

$_{4}^{\dagger}$Weeks after storage of soil treated with indaziflam
‡ Standard error comparing dose rates and soil storage time after indaziflam application

Table 4. Residual effect of different concentrations of indaziflam on melon (cm) seedlings at 0 to 4 weeks after treatment in 2016 and 2017

Dose rate (kg a.i/ha-^{1})	2016					2017				
	0	**1**	**2**	**3**	**4**[†]	**0**	**1**	**2**	**3**	**4**
0.000	14.380	13.858	17.421	18.243	38.210	14.290	13.807	17.403	18.282	38.443
0.004	4.157	12.403	12.427	12.411	36.003	4.201	12.389	12.404	12.432	36.373
0.009	2.763	10.331	10.660	11.541	30.700	2.600	10.583	10.366	11.192	30.733
0.013	1.320	10.014	10.363	10.148	37.120	2.214	10.329	10.171	11.500	37.593
0.018	1.875	10.070	10.179	10.069	36.637	2.189	10.055	10.007	10.074	36.630
0.020	1.143	6.783	7.920	8.863	19.020	1.804	6.780	7.877	8.861	19.110
0.027	1.375	3.400	7..075	7.604	13.907	1.726	3.403	8.092	9.420	13.983
0.031	1.265	3.400	4.603	6.756	12.650	1.459	3.237	3.504	7.805	12.417
0.036	0.670	3.214	3.320	3.383	10.440	0.668	3.007	3.324	3.389	10.480
0.040	0.643	3.007	3.310	3.546	10.430	0.639	3.017	3.308	3.472	10.250
0.045	0.631	3.009	3.307	3.356	10.250	0.607	0.821	0.862	1.703	2.823
[‡]SED (interaction)			0.1133					0.1909		

[†]Weeks after storage of soil treated with indaziflam

[‡] Standard error comparing dose rates and soil storage time after indaziflam application

Table 5. Residual effect of different concentrations of indaziflam on maize height (cm) seedlings at 0 to 4 weeks after treatment in 2016 and 2017.

Dose rate (kga.i/ha-^{1})	2016					2017				
	0	1	2	3	4[†]	0	1	2	3	4
0.000	14.396	15.196	17.430	18.313	38.137	14.400	14.897	17.450	18.240	38.12
0.004	4.200	12.460	12.449	12.480	36.177	4.210	12.477	12.470	12.480	36.10
0.009	2.758	10.640	11.340	11.564	30.777	2.783	10.537	11.367	11.570	30.81
0.013	1.331	10.359	10.476	10.957	37.133	1.330	10.340	10.480	10.980	37.15
0.018	2.081	10.532	10.559	10.647	36.677	1.763	10.537	10.553	10.673	36.69
0.020	2.010	6.745	8.484	8.867	19.027	1.580	6.767	8.470	8.870	19.03
0.027	1.720	3,402	8.038	8.607	13.557	1.740	3.403	8.020	8.610	13.98
0.031	1.450	1.534	1.593	6.758	12.707	1.457	1.530	1.597	6.793	12.79
0.036	0.668	1.038	1.324	2.189	10.523	0.670	1.053	1.307	2.190	10.56
0.040	0.637	1.028	1.311	2.266	10.240	0.640	1.030	1.300	2.310	10.28
0.045	0.601	0.685	0.904	1.752	2.953	0.600	0.680	0.910	1.780	2.98
[‡]SED (interaction)			0.1226					0.1227		

$_{4}$[†]Weeks after storage of soil treated with indaziflam

[‡] Standard error comparing dose rates and soil storage time after indaziflam application

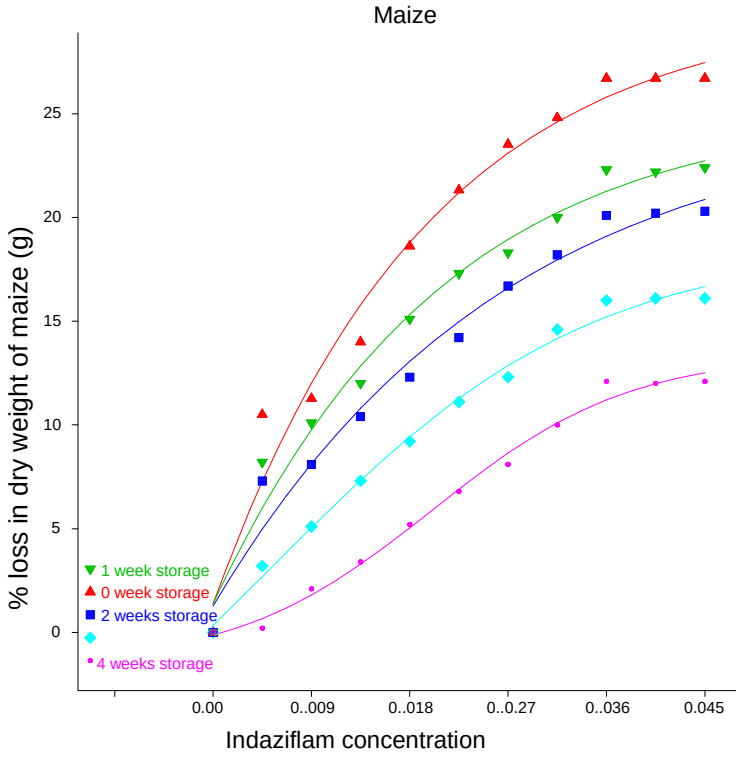

Fig. 1. The fitted dry weight of maize seedlings at various storage time at different herbicide concentrations The regression model is y=a+c/(1+exp(-b*(x-m))) For week 0: a =-444, b=242, c =474, m = -0.0113 and x = herbicide concentrations. For week 1 a =-213, b = 236, c =238, m = -0.0093 and x = herbicide concentrations applied. For week 2: a = -416, b =178, c =441, m =-0.016 and x = herbicide concentrations applied. For week 3: a = -12.3, b =308, c =30.9, m =0.00120 and x = herbicide concentrations applied. For week 4: a =- 1.60, b =498, c =15.01, m =0.004461 and x = herbicide concentrations applied. r^2 =0.993, S: E =0. 491 P>0.001

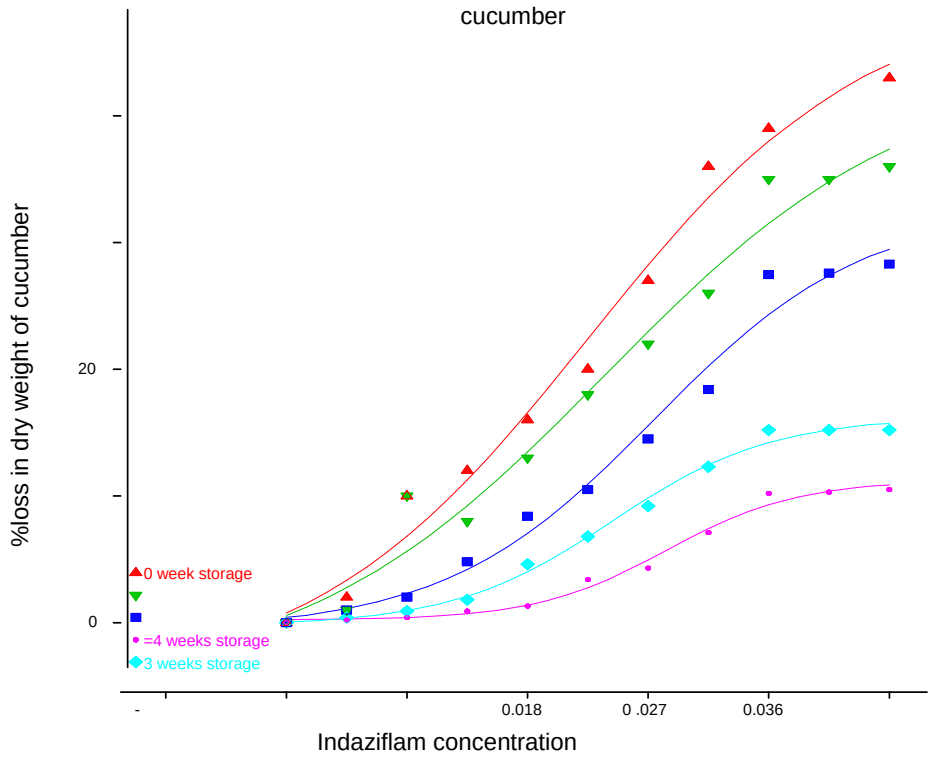

Fig 2. Fitted dry weight of cucumber seedlings at various storage time at different herbicideconcentration The regression model is y=a+c/(1+exp(-b*(x-m))).For week 0: a =-415, b=0.71, c =434, m = -3.4 and x = type of herbicide applied. For week 4: a =-0.002, b = 7, c =5, m = 2.08 and x = type of herbicide applied. For week 8: a = -3.1, b =0.415, c =6.8, m =1.56 and x = type of herbicide applied. For week 12: a = -0.06, b =0.7, c =0.24, m =3.0 and x = type of herbicide applied. For week 16: a = 0.00, b =3, c =0.023, m =4.1 and x = type of herbicide applied. r^2 =0.993, S: E =0. 491 P>0.001

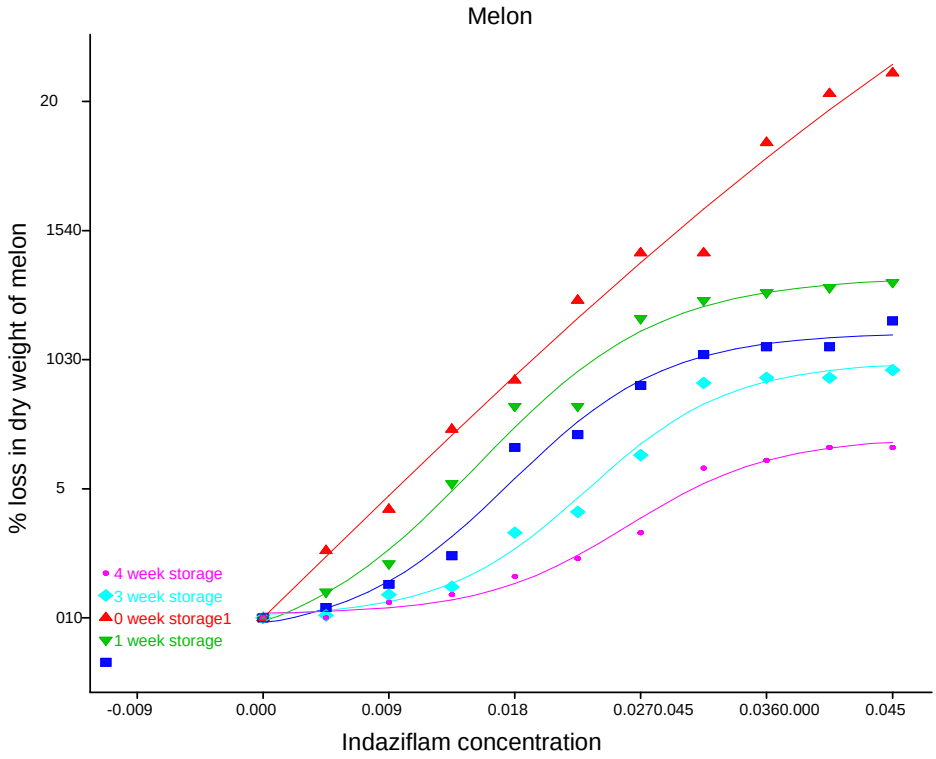

Fig 3. Fitted dry weight of melon seedlings at various storage time at different herbicide concentration The regression model is y=a+c/(1+exp(-b*(x-**m**))**).** For week 0: a =-415, b=0.71, c =434, m = -3.4 and x = type of herbicide applied. For week 4: a =-0.002, b = 7, c =5, m = 2.08 and x = type of herbicide applied. For week 8: a = -3.1, b =0.415, c =6.8, m =1.56 and x = type of herbicide applied. For week 12: a = -0.06, b =0.7, c =0.24, m =3.0 and x = type of herbicide applied. For week 16: a = 0.00, b =3, c =0.023, m =4.1 and x = type of herbicide applied. r^2 =0.993, S: E =0. 491 P>0.001

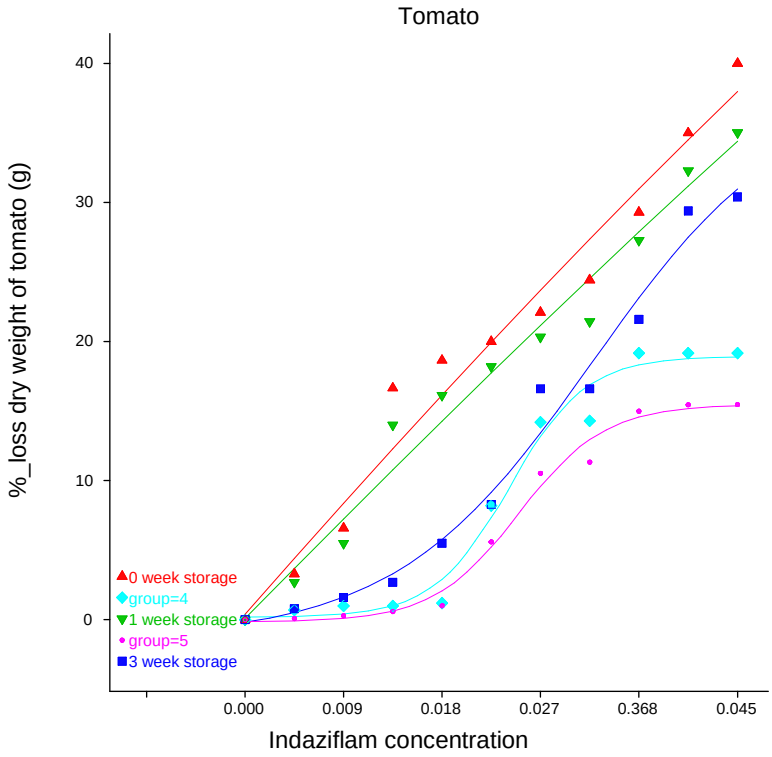

Fig 4. Fitted dry weight of tomato seedlings at various storage time at different herbicide contraction The regression model is y=a+c/(1+exp(-b*(x-m))). For week 0: a =-415, b=0.71, c =434, m = -3.4 and x = type of herbicide applied. For week 4: a =-0.002, b = 7, c =5, m = 2.08 and x = type of herbicide applied. For week 8: a = -3.1, b =0.415, c =6.8, m =1.56 and x = type of herbicide applied. For week 12: a = -0.06, b =0.7, c =0.24, m =3.0 and x = type of herbicide applied. For week 16: a = 0.00, b =3, c =0.023, m =4.1 and x = type of herbicide applied. r^2 =0.993, S: E =0. 491 P>0.001

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