

## Comparative Efficacy of Chemical Hybridising Agents as Seed and Foliar Spray Treatments on Wheat

RAJENDRA KUMAR, C. DEVAKUMAR, S.M.S. TOMAR<sup>1</sup> AND ASHOK GAUR<sup>2</sup>

Division of Agricultural Chemicals  
Indian Agricultural Research Institute, New Delhi- 110012  
cdevakumar@yahoo.com

**ABSTRACT** Chemical hybridizing agents (CHAs) help in "two line" heterosis breeding of wheat. Eight chemicals viz five *N*-acyl anilines and three analogues of *N*-aryl-5-carboxy-4, 6-dimethyl-1, 2-dihydroprid-2-ones were evaluated as CHAs both by foliar spray treatment at premeiotic stage as well as seed treatment. In general, foliar spray showed higher activity compared to seed treatment. The highest activity of 99.9 per cent at 1500 ppm foliar spray treatment was shown by ethyl-4-flouro oxanilate which was closely followed by 5-carboxy-*N*-(4-bromophenyl)-4, 6-dimethyl-1, 2-dihydroprid-2-one with 99.4 per cent male sterility. Ethyl-4-cyano oxanilate caused the highest male sterility of 65.3 per cent when seeds were treated at 1 per cent (w/w) of seeds. Even though none of the test chemicals has caused very high induction of male sterility as seed treatment, the very fact that the activity could be induced through seed treatment leads to two conclusions. First, the efficacy of the CHA can be persistent in the life cycle of the plant. Second, the seed treatment offers an alternative way for inducing male sterility.

**Keywords:** Chemical hybridizing agents, wheat, seed dressing, foliar spray, male sterility

Chemical Hybridizing Agents (CHAs) facilitate cross breeding in the plant species with perfect flower by sterilizing the male sex cells or by interrupting microsporogenesis to prevent self pollination and to promote fertilization by an outside pollen source [1]. CHAs are important in heterosis breeding of small cereal grain crop like wheat for which mechanical emasculation is impractical due to the small size and close proximity of male and female reproductive organs.

In a programme of design and development of CHAs, we have undertaken the synthesis and screening of several *N*-acylanilines, amino acid analogues and pyridones at the Indian Agricultural Research Institute. Encouraging results have been obtained with field trials of a few *N*-acylanilines on rice [2, 3], wheat [4, 5, 6] and chickpea [7]. Quantitative structure-activity relationship analysis of oxanilates revealed a direct relationship between Swain-Lupton field constant and molecular masses but an inverse relationship between molar refractivity, position of the aromatic substituents and Swain -Lupton resonance constant in influencing

the bioactivity [8]. The aim of the all above studies was to induce male sterility by foliar spray of the plant at specific stage of plant growth. In this communication we report the relative efficacy of the selected *N*-acyl anilines and pyridones as seed treatment at the time of sowing in wheat.

### MATERIALS AND METHODS

Eight promising CHAs of proven male sterlant activity on wheat after foliar spray treatment at premeiotic stage when the length of panicle of the first node was 7-8 mm were tested as seed treatment. Five *N*-acyl anilines were synthesized by thermal condensation of diethyl oxalate with corresponding anilines in toluene. Three acid derivatives of *N*-aryl-5-carboxy-4, 6-dimethyl-1, 2-dihydroprid-2-one were synthesized by thermal condensation of ethyl isodehydroacetate with corresponding substituted anilines in aqueous acetic acid (50%) for 12-18 hrs followed by alkali hydrolysis of carboethoxy intermediate for 6 hrs in methanolic NaOH (5%). The sodium salt so obtained was converted into

1. Division of Genetics

2. Division of Seed Science and Technology

acids by neutralization with dilute HCl. The course of the reaction was monitored by TLC and HPLC. The structures of the compound were confirmed by melting point and NMR analysis.

The high yielding variety of bread wheat (*Triticum aestivum* L.) PBW 343 was chosen for evaluation of male sterility through seed treatment. The seed dressing of test compounds was done at 0.5 and 1% (w/w) of seed using kaolin as an inert carrier. Stock solutions (25%) of test-compound were prepared in acetone. To the test-compound in acetone (1 ml) requisite quantities of kaolin (200mg) and one drop of gum acacia in water (20%) were added and mixed thoroughly. Then the required quantities of wheat grains (25 g) were added and mixed intermittently to allow the evaporation of acetone. The wheat grains were air-dried overnight. The coated seeds along with control were sown in IARI farm following a randomized block design in three replicates in rabi 2002-03. A parallel experiment for foliar spray treatment was also conducted in randomized block design with three replicates. The test compounds were formulated as 15% EC in cyclohexanone containing Tween 80 (5%) as emulsifier. The crop was sprayed at 1000 and 1500 ppm at the premeiotic stage of plant growth when the length of the spike emerging out from the first node was 7-8 mm.

Ten spikes along with control in each replicate were bagged immediately after emergence and the seeds set in each spike were counted at harvest in foliar spray as well as seed dressing treatments. The per cent male sterility was computed as per cent inhibition of seed set in bagged spikes of treated plants using the formula:

$$\text{Per cent sterility} = (S_c - S_t) / S_c \times 100$$

Where,  $S_c$  = seeds per spike in control plants

$S_t$  = seeds per spike in treated bagged plants

## RESULTS AND DISCUSSION

The effect of foliar spray and seed dressing treatment of eight synthetic compounds on spikelet fertility of wheat variety PBW 343 at two concentrations is shown in Table 1.

### Foliar spray application

The foliar spray application at premeiotic stage was able to induce partial to nearly complete male sterility and generally showed higher activity compared to seed treatment. It may be seen from the table that the maximum male sterility was induced by ethyl-4-fluoro oxanilate, which differed significantly over other oxanilates and pyridones. It

Table 1. Male sterility induced by tests CHAs as foliar spray and seed treatment on wheat variety PBW343

Sr No	Test Compound	Spray Treatment		Seed Treatment	
		%Dose	%Sterility	%Dose	%Sterility
1	Ethyl-4-cyano oxanilate	0.10	82.49	0.50	18.59
		0.15	93.85	1.00	65.27
2	Ethyl-4-fluoro oxanilate	0.10	99.54	0.50	8.63
		0.15	99.97	1.00	17.96
3	Ethyl-4-bromo oxanilate	0.10	88.69	0.50	1.20
		0.15	97.14	1.00	27.54
4	Ethyl-4- trifluoromethyl oxanilate	0.10	86.18	0.50	21.56
		0.15	95.97	1.00	32.34
5	Ethyl-4-chloro oxanilate	0.10	49.67	0.50	4.79
		0.15	72.09	1.00	30.50
6	5-carboxy-N-(4-fluorophenyl)-4, 6-dimethyl-1, 2-dihydroprid-2-one	0.10	3.70	0.50	29.90
		0.15	5.57	1.00	52.10
7	5-carboxy-N-(4-bromophenyl)-4, 6-dimethyl-1, 2-dihydroprid-2-one	0.10	96.30	0.50	19.76
		0.15	99.45	1.00	38.82
8	Na N-(4-fluorophenyl)-4, 6-dimethyl-1, 2-dihydroprid-2-one-5-carboxylate	0.10	94.10	0.50	1.80
		0.15	98.98	1.00	16.77
CD (P=0.05)			0.66	12.98	

induced 99.5 and 99.9% male sterility at 1000 and 1500 ppm concentrations, respectively. It was closely followed by 5-carboxy-N-(4-bromophenyl)-4, 6-dimethyl-1, 2-dihydroprid-2-one with 96.3 and 99.4 per cent male sterility at 1000 and 1500 ppm concentrations, respectively. Sodium salt of N-(4-fluorophenyl) - 4, 6-dimethyl- 1, 2-dihydroprid-2-ones-5-carboxylate, ethyl-4-bromo oxanilate and ethyl-4-trifluoromethyl oxanilate were also found to be excellent in inducing 98.93, 97.14 and 95.17 per cent male sterility at 1500 ppm concentrations respectively. 5-carboxy-N-(4-fluorophenyl)-4, 6-dimethyl-1, 2-dihydroprid-2-one showed the lowest activity at both the concentrations.

#### Seed treatment

It was observed that ethyl-4-cyano oxanilate and 5-carboxy-N-(4-fluorophenyl)-4, 6-dimethyl-1, 2-dihydroprid-2-one caused 65.27 and 52.10 per cent induction of male sterility at 1% dosage. All other compounds were found to exhibit very low degree of male sterility at both concentrations of seed dressing.

The ethyl-4-fluoro oxanilate gave the highest activity when used as foliar spray whereas ethyl-4-cyano oxanilate was superior as seed treatment. This is the first report on the assay of CHAs as seed treatment.

Chemical induction of male sterility is generally achieved by foliar spray of CHAs [2-8]. However this has certain disadvantages [1]. The efficacy is dependent on growth stage and weather conditions such as rain, cloudy climate etc. have a drastic influence. It is not suitable for non-synchronous flowering crops. Late tillering is a problem as the late tillers would not be sterile. Even though none of the test chemicals has caused high induction of male sterility as seed treatment, the very fact that the activity could be induced through seed treatment

leads to two conclusions. First, the activity of the CHA can be persistent through the life cycle of the plant. Second, the seed treatment offers an alternative way for inducing male sterility.

It is therefore felt that seed treatment could be an alternative and efforts should be made to develop chemicals which produce high degree of male sterility. In this context, the present work is a way forward.

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