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A Review :

Monitoring of herbicide (atratat 50W) toxicity by using pollen as indicators - Pollen of five cultivars of *Petunlia axillaris* BSP.: Further evidence of a criticism of Berg (1973), Brandt (1974), Rasmussan (1977), Navara, Horvath and Kaleta (1978), Mhatre (1980 - Ph.D. Thesis), Mhatre, Chaphekar, Ramani Rao, Patil, Haldar (1980), Shetye (1982 - Ph.D. Thesis) and Giridhar (1984 - Ph.D. Thesis) - A Critical Review

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Germinability of pollen of F series of white-flowered and F-24 series of light-violet-, pink-, violet-, white-flowered and F-48 series of white-flowered cultivars of *Petunia axillaris* was suppressed even by the lowest concentration (10^{-17} mg/ml) of atratat 50W tried

Key words : Physiology of Pollen, Palylnology, Toxicology, Environmental Sciences,

Extensive use of herbicides leaves behind residues which contaminate our environment. It is primarily needed to work out some simple system for the evaluation of the toxicity of herbicides.

Pollen of successive flowers (*viz.* F, F-24, F-48, F-72 series *i.e.* open flowers and the flower buds which require 24, 48, 72 hours to open respectively) of 5 cultivars of *Petunia axillaris* BSP. *e.g.* light-violet-, pink-, violet-, white- and white-violet-flowered cultivars were collected at the stage of the dehiscence of anthers in the open flowers. Germination of pollen grains of successive flowers was studied by standing-drop technique in the optimum concentrations of sucrose (acts as control) as well as in the optimum concentrations of sucrose supplemented by the different concentrations (10^{-17}-10^{-2}-10^{-3}, 1, 5, 10, 20-20-100 mg/ml) of atratat 50W (Table 1). The cultures were then transferred to a moist filter chamber, stored at room temperature (21.9-32.2°C) having RH 58% and in diffuse laboratory light. Observations were recorded 24 hours after incubation. For each experiment a random count of 200 grains was made to determine the percentage of pollen germination. For measurement of length of pollen tubes, 50 tubes were selected randomly and measured at a magnification of 100x.

Potentiality of pollen germinability was noted in F and F-24 series of all the 5 cultivars of *Petunia axillaris* and in F-48 series of white-flowered cultivar of *P. axillaris*. Thus the potentiality of pollen germinability in *P. axillaris* was recorded in 11 out of 20 series investigated (Table 1). Potentiality of the germinability of pollen is noted only in F series of pink- and white-flowered cultivars of *Nerium odorum*. Both of them are single-flowered cultivars (Salgare, 1983-Ph.D.Thesis). Potentiality of the germinability of pollen was recorded in F and F-24 series of *Physalis minima* and *Solanum xanthocarpum* (Ram Indar, 1981-M.Sc.Thesis), in red-flowered (double-flowered) cultivar of *N. odorum* and in white-flowered cultivar of *Catharanthus roseus* (Salgare, 1983) and in all the cultivars (light-violet-, pink-, violet- and white-violet-flowered cultivars) of *Petunia axillaris* except for white-flowered cultivar (Salgare, 1986a-Ph.D.Thesis-Table 1). Pollen germination *in vitro* culture of sucrose was noted in F, F-24 and F-48 series of *Brunfelsia americana* and in violet-flowered form of *Datura fastuosa* (Ram Indar, 1981) and in white-flowered cultivar of *P. axillaris* (Salgare, 1986a). However, it was the pollen of white-flowered form of *D. fastuosa* (Ram Indar, 1981) and pink-flowered cultivar of *C. roseus* (Salgare, 1983)

showed their germination *in vitro* culture of sucrose in all the 4 series (F, F-24, F-48, F-72 series) investigated. Potentiality of the germinability of pollen in all the 4 series investigated was also noted by Salgare (1986d-D.Sc.Thesis) in 3 Leguminous crops *viz. Cyamopsis tetragonoloba* Taub. var. Pusa Navbahar – gawar, *Phaseolus aureus* Roxb. var. J-781- mung and *Phaseolus mungo* Roxb. var. T-9- urid. Theresa Sebastian (1987-Ph.D.Thesis) observed the germination of pollen of one of the Leguminous crops *i.e. Vigna mungo* (L.) Hepper Type 9, of Uttar Pradesh in all the 4 series investigated *in vitro* culture of sucrose. Suwarna Gawde (1988-Ph.D.Thesis) noted the germinability of pollen of 2 Leguminous crops *viz. Vigna unguiculata* (L.) Walp. var. Pusa Barsati – cowpea and *Vigna radiata* (L.) Wilczek. var. Pusa Baisakhi of Delhi in all the 4 series investigated. Johri and Chhaya Roy Chowdhury (1957) stated that in *Citrullus colocynthis,* where pollen grains 'mostly remained attached in tetrads', satisfactory germination is observed.

Salgare (1983) observed the germination of pollen of F-72 series of pink-flowered cultivar of *Catharanthus roseus in vitro* culture of sucrose. Trisa Palathingal (1990-M.Phil.Thesis) stated that the pollen of F-72 series of pink-flowered cultivar of *Catharanthus roseus* did not germinate in Brewbaker and Kwack's (1963) culture medium. This proves that the culture medium is also having the bearing on the germination of pollen. This points out that Brewbaker and Kwack's (1963) culture medium is not ideal for the pollen culture.

Even the lowest concentration (10^{-17} mg/ml) of atratat 50W tried suppressed the germinability of pollen in 2, 9, 6 series of Apocynaceae (Salgare, 1983), *Petunia grandiflora* (Sharma, 1984-Ph.D.Thesis) and *Petunia axillaris* (Salgare, 1986, Table 1) respectively. It should be pointed out that even the lowest concentration (10^{-17} mg/ml) of atratat 50W tried suppressed the germinability of pollen of F-24 series of red-flowered cultivar of *Neriulm odorum* and F-72 series of pink-flowered cultivar of *Catharanthus roseus* (Salgare, 1983). Sharma (Sharma, 1984) stated that the germinability of pollen of F series of duet, sonata and F-24 series of pink cascade, white cascade, duet and sonata and F-48 series of all the 3 cascades of the cultivars of *P. grandiflora* was prevented even by the lowest concentration (10^{-17} mg/ml) of atratat 50W tried. Germinability of pollen of F series of white-flowered and F-24 series of light-violet-, pink-, violet-, white-flowered and F-48 series of white-flowered cultivars of *P. axillaris* was suppressed even by the lowest concentration (10^{-17} mg/ml) of atratat 50W tried (Table 1). This proves that the pollen of the said series are highly sensitive and acts as an ideal indicators of pollution. Thus it is confirmed that the pollen development and activity are more sensitive indicators of adverse factors in the botanical environment and the use of an entire vascular plant (Berg,1973; Brandt, 1974; Vick and Bevan, 1976; Rasmussan, 1977; Navara, Horvath and Kaleta, 1978; Mhatre, 1980-Ph.D.Thesis; Mhatre, Chaphekar, Ramani Rao, Patil, Haldar, 1980; Shetye, 1982-Ph.D.Thesis and Giridhar, 1984-Ph.D.Thesis) as an indicator of pollution is a very crud method and rather a wrong choice. There is no evidence of any entire vascular plant exhibiting this much degree of sensitivity. This is confirmed in the present critical review (Table 1) and was also proved earlier by Salgare (1983, 84, 85a-c, 86a-d, 2000, 1a-b, 05a- c, 06), Salgare and Theresa Sebastian (1986), Salgare and Phunguskar (2002), Salgare and Sanju Singh (2002) and Salgare and Sanchita Pathak (2005) and Salgare's Research Group (Ram Indar, 1981; Sharma, 1984; Singh, 1985; Theresa Sebastian, 1987; Suwarna Gawde, 1988 and Trisa Palathingal, 1990) also supports the present findings.

Inhibition in the germination of pollen was caused by atratat 50W in 7, 4, 5 series of Apocynaceae (Salgare, 1983), *Petunia grandiflora* (Sharma, 1984) and *Petunia axillaris* (Table 1) respectively.

The widest range of concentrations of atratat 50W found to be 10^{-17}- 40, 10^{-17}-100, 10^{-17}-1 mg/ml which inhibited the germination of pollen of *Petunia axillaris* (in F series of pink-flowered cultivar) (Salgare, 1986a-Table 1), Apocynaceae (in F series of pink-flowered cultivar of *Nerium odorum*) (Salgare, 1983) and *Petunia grandiflora* (in F series of pink cascade) (Sharma, 1984) respectively.

Sub-toxic concentration of atratat 50W caused as high as 94.29% and 96.55% inhibition in the pollen germination of *P. axillaris* (in F series of light-violet-flowered cultivar) (Table 1) and *P. grandiflora* (in F series of pink cascade) (Sharma, 1984) respectively.

Ratio between the series and inhibition caused by atratat 50W (in sub-toxic concentration) in the germination of pollen is as (Inhibition is represented in the form of percentage):

F:F-24:F-48:F-72 = 90.88:92.86:00.00:00.00 in *Petunia axillaris* (Table 1-Salgare, 1986a)

F:F-24:F-48:F-72 = 70.00:00.00:00.00:00.00 in Apocynaceae (Salgare, 1983)

F:F-24:F-48:F-72 = 87.10:91.67:00.00:00.00 in *Petunia grandiflora* (Sharma, 1984)

This shows that atratat 50W caused maximum inhibition in the germination of pollen of F series of Apocynaceae (Salgare, 1983) and of F-24 series of

Petunia grandiflora (Sharma, 1984) and *Petunia axillaris* (Table 1)(Salgare, 1986a).

Atrataf 50W stimulated the pollen tube growth in 5, 0, 0 series of Apocynaceae (Salgare, 1983), *Petunia grandiflora* (Sharma, 1984) and *Petunia axillaris* (Salgare, 1986a) respectively.

Atrataf 50W inhibited the pollen tube growth in 7, 4, 5 series of Apocynaceae (Salgare, 1983), *Petunia grandiflora* (Sharma, 1984) and *Petunia axillaries* (Salgare,1986a) respectively.

The widest range of the concentrations of atrataf 50W tried, found to be 10^{-17}- 40, 10^{-17}-100, 10^{-17}-1 mg/ml which inhibited the pollen tulbe growth of *Petunia axillaris* (in F series of pink-flowered cultivar) (Salgare, 1986a-Table 1), Apocynaceae (in F series of pink-flowered cultivar of *Nerium odorum* and *Catharanthus roseus*)(Salgare, 1983) and *Petunia grandiflora* (in F series of pink cascade) (Sharma, 1984) respectively.

Sub-toxic concentration of atrataf 50W caused as high as 97.14% inhibition in the pollen tube growth of *Petunia axillaris* (in F series of white-violet-flowered cultivar) (Salgare, 1986a-Table 1). However, the maximum inhibition in the pollen tube growth (94.74%) was reported by Sharma (1984) in *Petunia grandiflora* (in F series of pink cascade).

Ratio between the series and inhibition caused by atrataf 50W (in sub-toxic concentration) in the pollen tube growth is as (Inhibition is represented in the form of percentage):

F:F-24:F-48:F-72 = 76.37:96.00:00.00:00.00 in *Petunia axillaris* (Table 1-Salgare, 1986a)

F:F-24:F-48:F-72 = 87.40:00.00:00.00:00.00 in Apocynaceae (Salgare, 1983)

F:F-24:F-48:F-72 = 91.98:94.74:00.00:00.00 in *Petunia grandiflora* (Sharma, 1984)

This shows that atrataf 50W caused maximum inhibition in the pollen tube growth of F series of Apocynaceae (Salgare, 1983) and in F-24 series of *Petunia grandiflora* (Sharma, 1984) as well as *Petunia axillaris* (Salgare, 1986a-Table 1).

Tube length *in vitro* culture (sucrose + atrataf 50W) of atrataf 50W (in sub-toxic concentration) is 0.03% (in) in *Petunia axillaris* of the tube length found *in vivo* is the longest of all the cultivars investigated (Table 1).

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