FULL LENGTH RESEARCH ARTICLE

THE EFFECTS OF DIETHYLSULPHATE ON SOME QUANTITATIVE TRAITS OF TOMATO (*Lycopersicon esculentum* Mill)

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ABSTRACT
Seeds of three varieties of tomato (*Lycopersicon esculentum* Mill) T106, T244 and T420 obtained from the Institute for Agricultural Research, A.B.U Zaria, were treated with Diethylsulphate (DES) at concentrations of 10, 15 and 30 mM with the aim of determining the effective concentration of the mutagen capable of inducing beneficial mutants in tomato. Highly significant differences (P<0.01) were observed in the varieties and treatment with respect to the traits studied. Seed germination, root length, height at maturity, number of branches per plants, and number of fruits per plant decreased with increase in mutagen concentration. Interactions between variety and concentrations were also highly significant (P<0.01) with respect to germination percentage, root length, seedling survival, height at maturity and number of branches per plant. In general variety T106 showed maximum performance when compared to T244 and T420. The highest number of fruits among the mutants was recorded in variety T244 at 30mM. DES therefore is an effective agent in tomato mutagenesis and 30 mM concentration could be used to induce variability that could be utilized for the improvement of tomato and other related economic crops.

Keywords: Diethylsulphate, chemical mutagens, mutagenesis, tomato, economic crops.

INTRODUCTION
Tomato is an important economic crop that suffers from diseases, pest infestations and adverse effects of environmental stress, which affect the total production. Current research on tomato focuses on improving yield, flavour and resistance to diseases (Encarta 2005).

Mutations either spontaneous or artificially induced, both in seeds and vegetatively propagated crops are of scientific and commercial interest. Mutations are the tools the geneticists use to study the nature and function of genes which are the building blocks and basis of plant growth and development, thus producing raw materials for genetic improvement of economic crops (Adamu *et al*. 2004). Various mutagenic agents were used to induce favorable mutations at high frequency; the use of ionizing radiation, such as x-rays, gamma-rays and neutrons as well as chemical mutagens for inducing variation is well established (Ahloowalia and Maluszynski 2001). The most effective chemical mutagens are alkylating agents especially ethylmathane sulphonate (EMS), N – methyl-N- nitro – guanidine (NE), Diethylsulphate (DES) and Dimethylsulphate (DMS) (Coe & Neuffer 1977; Freeze 1971; Holleander 1973; Khalitkar & Bhatia 1975; Ricardo & Ando 1998). Compared with ionizing radiation, chemical mutagens show proportionately smaller aberration and fewer lethals are induced (Mashenkov 1986).

The present work reports the effectiveness of DES in inducing beneficial mutants in tomato.

MATERIALS AND METHODS
Acquisition and preparation of seeds: Air dried seeds of tomato (T106, T244 and T420) obtained from the Horticultural section of Institute of Agricultural Research, A.B.U Zaria, were soaked in freshly prepared DES at concentrations of 15, 20 and 30 mM in 0.1M phosphate buffer pH 6 for 4 hours. The control seeds were soaked in buffer solution pH 6. The treatments were periodically agitated at room temperature. Thereafter, seeds were thoroughly rinsed with tap water to remove excess mutagens.

Planting of seeds: The seeds were planted in the field and laboratory. In the laboratory, sowing involved planting treated seeds in lunch boxes. Planting in the field involved the use of pots to obtain the seedlings which were later transplanted to beds after 4 weeks as the M 1 generation. Each treatment was replicated five times in a completely randomized design (CRD).

Parameters measured: Data were collected on seed germination percentage, seedling height, number of leaves per seedling, seedling survival, height at maturity, number of branches per plant, delayed development /maturity and number of fruits (yield) per plant.

The data obtained were analyzed using analysis of variance (ANOVA) and where significant differences were found, Duncan's multiple range test (DMRT) was used to separate the means.

RESULTS
The results of the study indicated highly significant differences (P<0.01) for germination percentage, root length, seedling survival, height at maturity, number of branches per plant, flowering /maturity and yield per plant in the three varieties of tomato exposed to DES in the M1 generation (Table 1). There was general decrease in germination percentages, seedling height, root length, number of leaves/seedling, seedling survival, height at maturity and number of branches per plant with increase in mutagen concentration. The number of fruits per plants however increases with increase in mutagen concentration in T244 (Table 2).

The interaction between variety and treatment was highly significant (P<0.01) for germination percentage, root length, seedling survival, height at maturity and branches per plant (Table 3). Furthermore, result shows highly significant differences (P<0.01) in the performance of the three varieties with respect to the traits studied. In general, variety T106 shows better performance when compared to T244 and T420 (Table4). The three different concentrations of the chemical mutagens were effective in inducing mutations and the highest concentration (30 mM) been the most effective in the varieties studied (Table 5).

TABLE 1: MEAN SQUARE OF THE EFFECTS OF DIETHYLSULPHATE ON TOMATO (*Lycopersicon esculentum* Mill]

Source of Variation	df	Germination % 5days	Germination % 8days	Seedling height (cm)	Root length (cm)	Number of leaves/ seedling	% seedling survival	Height at maturity (cm)	Number of branches/ plant	50% flowering (days)	Number of fruits/plant
Treatment	15	3145.44**	2226.67**	220.41ns	103.92**	6186.19ns	22.71**	793.63**	37.26**	15.59.**	1481.61**
Error	44	24.05	17.83	267.36	5.27	5778.63	2.25	85.57	2.75	1.59	120.99

*=significant (P<0.05) ns = non-significant **=significant (<0.01)

TABLE 2: MUTAGENIC EFFECTS OF DIETHYLSULPHATE ON TOMATO (*Lycopersicon esculentum* Mill)

Variety	Treatment	Germination % 5days	Germination % 8days	Seedling height (cm)	Root length (cm)	Number of leaves/ seedling	% seedling survival	Height at maturity (cm)	Number of branches/plant	50% flowering (days)	Number of fruits/plant
T106	0mM	70.60^{b}	92.40^{a}	15.88	8.04^{b}	14.00	90.00^{a}	67.60^{a}	8.00^{b}	86.40^{c}	63.00^{a}
	15mM	40.00^{c}	89.80^{a}	10.88	10.12^{b}	9.00	69.80^{c}	52.36^{a}	5.00^{e}	87.20^{b}	13.00^{c}
	20mM	89.80^{a}	91.80^{a}	21.10	20.80^{a}	15.00	69.70^{c}	64.36^{a}	10.00^{b}	89.00^{b}	10.00^{c}
	30mM	68.40^{b}	76.20^{b}	16.42	18.42^{a}	12.00	61.00^{d}	58.36^{a}	13.00^{a}	89.60^{b}	17.00^{c}
T244	0mM	2.00^{f}	72.20^{b}	13.02	6.88^{c}	10.00	70.40^{c}	53.80^{c}	14.00^{a}	86.20^{c}	52.00^{a}
	15mM	12.00^{e}	45.80^{d}	12.14	4.36^{d}	7.00	58.00^{e}	32.80^{c}	9.00^{b}	89.20^{b}	4.00^{d}
	20mM	10.20^{e}	25.60^{f}	10.68	5.84^{c}	7.00	65.60^{f}	24.40^{d}	7.00^{c}	90.60^{a}	6.00^{d}
	30mM	4.00^{f}	24.80^{f}	9.66	6.18^{c}	7.00	65.00^{f}	31.00^{c}	7.00^{c}	91.60^{a}	23.00^{b}
T420	0mM	23.30^{d}	77.40^{b}	22.24	6.34^{c}	13.00	82.00^{b}	41.36^{b}	6.00^{d}	86.00^{c}	44.00^{a}
	15mM	19.80^{d}	53.20^{d}	17.36	4.10^{d}	13.00	61.00^{g}	51.60^{b}	6.00^{d}	86.20^{c}	14.00^{c}
	20mM	23.20^{d}	62.00^{c}	33.50	5.22^{d}	11.00	61.00^{g}	39.20^{b}	6.00^{d}	90.20^{a}	13.00^{c}
	30mM	13.00^{e}	40.20^{c}	16.72	3.22^{d}	10.00	50.20^{h}	30.60^{c}	4.00^{e}	90.40^{a}	6.00^{d}

Means with the same letter are not significantly different

TABLE3: MEAN SQUARE VALUES OF EFFECTS OF DIETHYLSULPHATE ON TOMATO (*Lycopersion escolentum* Mill)

Source of variation	df	Germination % 5 days	Germination % 8 days	Seedling height (cm)	Root length (cm)	Number of leaves/ seedling	% seedling survival	Height at maturity (cm)	Number of branches/ plant	50% flowering (days)	Number of fruits/plant
Replication	4	6.73^{ns}	31.33^{ns}	217.25^{ns}	2.13^{ns}	6076.40^{ns}	1.82^{ns}	153.34^{ns}	3.58^{ns}	12.44**	194.11^{ns}
Variety	2	20066.67**	10630.02**	618.59^{ns}	496.49**	3211.41^{ns}	98.13**	3532.22**	99.26**	4.12*	237.22^{ns}
Treatment	3	785.44**	2877.64**	210.62^{ns}	56.33**	10492.99^{ns}	11.16*	572.36**	10.96*	51.35**	67467.44**
Variety x Treatment	3	777.54**	563.51**	94.69^{ns}	64.73**	5097.58^{ns}	17.27**	416.64**	52.64**	3.65^{ns}	261.82^{ns}
Error	44	24.05	17.83	267.36	5.27	5778.63	2.25	85.87	2.75	1.59	120.99

*= significant (P<0.05), **= significant (P<O.01) ns=non-significant

TABLE 4: MEAN PERFORMANCE OF THREE VARIETIES OF TOMATO (*Lycopersicon esculentum* Mill) TREATED WITH DIETHYLSULPHATE.

Variety	Germination % 5 days	Germination % 8 days	Seedling height (cm)	Root length (cm)	Number of leaves/ seedling	% seedling survival	Height at maturity (cm)	Number of branches/ plant	50% flowering (days)	Number of fruits/plant
T106	67.20^{a}	87.55^{a}	16.07	13.84^{a}	12.00	72.65^{b}	60.67^{a}	9.00^{a}	88.08^{b}	26.00^{a}
T244	7.10^{c}	42.10^{c}	11.38	5.82^{b}	8.00	63.75^{b}	35.50^{b}	9.00^{a}	88.90^{a}	22.00^{a}
T420	19.80^{b}	58.10^{b}	22.45	4.72^{b}	12.00	88.75^{a}	40.69^{b}	5.00^{b}	88.20^{ab}	19.00^{b}

Mean with the same letters within column are not significantly different.

TABLE 5: MUTAGENIC EFFECT OF DIETHYLSULPHATE ON THREE VARIETIES OF TOMATO (*Lycopersicon esculentum* Mill)

Treatment	Germination % 5 days	Germination % 8 days	Seedling Height (cm)	Root length (cm)	Number of leaves/ seedling	% seedling survival	Height at maturity (cm)	Number of branches/ plant	50% flowering (days)	Number of fruits/ plant
0mM	31.30^{b}	80.67^{a}	17.05	7.09^{bc}	12.00^{a}	114.40^{a}	54.25^{a}	9.00^{a}	86.20^{c}	53.00^{a}
15mM	25.70^{c}	62.80^{b}	13.46	6.19^{c}	10.00^{c}	62.23^{b}	45.59^{b}	7.00^{b}	87.53^{b}	10.00^{b}
20mM	41.90^{a}	59.80^{b}	21.76	10.62^{a}	11.00^{b}	65.47^{b}	42.65^{b}	8.00^{b}	89.60^{a}	10.00^{b}
30mM	27.10^{b}	47.07^{c}	14.23	8.61^{b}	10.00^{c}	57.40^{b}	39.99^{b}	8.00^{b}	90.20^{a}	12.00^{b}

Mean with the same letters within column are not significantly different.

DISCUSSION

Result from this study showed that treatment of tomato varieties with DES produce mutants that vary with respect to the traits studied. The decrease in seedling emergence, seedling height, root length, seedling survival is characteristic of the effect of mutagenic agents as reported by Adamu *et al*. (2002) when groundnut was treated with gamma rays. In the study of the effects of ionizing radiation on tomato, it was observed that these traits were dose dependent (Adamu *et al*. 2004). Similar result was obtained by Sheeba *et al*. (2005) when gamma rays and EMS were employed to treat sesame seed (*Sesamus indicum* L) germination, seedling survival, plant height and pollen sterility when significant reduction was observed with an increase in dosage levels of both mutagens. This also agreed with the findings of Sasi *et al*. (2005) on the effects of DES and EMS on Okra (*Abelmoscus esculentum* L. var.MDU-I), who observed that all plant mutant types registered lower yield compared to their parent Okra (control).

In general DES was efficient in increasing variability of the germination, root length, seedling survival, height at maturity and number of branches per plant in tomato. However seedling height and number of leaves per seedling were not affected. Also the varieties of tomato performed differently with the mutagen and T106 shows the highest response with respect to all the traits studied except in number of fruits per plant where T244 produced the highest number of fruit. Furthermore, 30 mM concentration of DES is the most effective concentration in tomato mutagenesis. DES at 30 mM could therefore be utilized to induce favorable mutations in tomato which will invariably increase the possibility of isolating beneficial mutants for further improvement of tomato production

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