# The Effects of Ethyl Methanesulphonate and Sodium Azide on Growth in 'G.G.Gerbing' azalea (Rhododendron) 

Meiping Cai', Qingxi Chen ${ }^{1}$, Mengmeng Gu², and Xiaoya Cai²

Fujian Agriculture and Foresty Univesity,College of Horticulture; ${ }^{2}$ Texas A\&M University, College of Agriculture and Life Sciences; meipingcai@tamu.edu; cox0246@163.com

## Introduction

$>$ Mutagens have become an important tool in generating new variability that can be used to improve yield and quality in many crops and ornamental plants.
> ethyl methanesulphonate (EMS), and sodium azide (SA) have been the most commonly used mutagens in plant breeding programs, primarily because of the fact that they do not disturb desirable attributes in plants.

## Objectives

The main objective of the experiment is to determine the effectiveness and optimum doses of chemical mutagens to induce the changes of growth and physiology responses in azalea.

## Materials and Methods

* 'G.G.Gerbing' azalea plants procured from Magnolia Gardens Nursery, Magnolia, TX,were transplanted in 6 inch pots filled with Berer growing media on 16 Dec. 2015 in an unshaded greenhouse at College Station, TX.
* The treatment, uniform, disease and pest free plants were selected. Each pot should contain 6 buds, and the extra buds were manually removed.
* Total of 126 plants were selected,the buds(was wrapped by the absorbent cotton) of these plants were treated with EMS and SA-solutions prepared in 0.1 M phosphate buffer adjusted to pH 7.4 and pH 3.0, respectively. The treatment concentrations, viz., $30 \mathrm{mM}, 60 \mathrm{mM}, 90$ $\mathrm{mM}, 120 \mathrm{mM}$ and 150 mM of EMS and $\mathbf{2 ~ m M , ~} 4$ $\mathrm{mM}, \mathbf{6 m M}, 8 \mathrm{mM}$ and 10 mM of SA.
\& During the establishment stage, plants were irrigated as needed using a nutrient solution containing $200 \mathrm{mg} \cdot \mathrm{L}-1$ 21N-7P-7K and reverse osmosis water.
*Environment: $40.9^{\circ} \mathrm{C}$ day $/ 24.2^{\circ} \mathrm{C}$ night; $24.9 \%$ RH; $22.6 \mathrm{~mol} \cdot \mathrm{~m}-2 \cdot \mathrm{~d}-1$ DLI.
* Measured parameters in Exps: Visual rating of buds, new buds height, new leaf number, aberrant leaf and chlorophyll content.


Firgure 1. The survial rate of EMS and SA treatmnet.


Figure 2. Responses of rating score among EMS and SA treatment solution.

| Mutagen | Time | h1 | h2 | h3 | h4 | 11 | 12 | 13 | 14 |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| EMS | 1 d | 0.6 b | 4.6 b | 7.8a | 9.6 a | 3.1a | 10.3a | 14.6a | 18.2a |
|  | 2 d | 0.9a | 5.9a | 8.8a | 10a | 3.6a | 11.3a | 15.9 | 18.6a |
|  | 3 d | 0.5b | 2.9c | 5.5b | 6.3b | 0.35a | 6.3b | 11.8 b | 17a |
| SA | 1 d | 1.16 | 6.5a | 10.4a | 20.1a | 3.4a | 10.8a | 15.5a | 11.7a |
|  | 2 d | 1.3a | 6.3a | 9.3 ab | 18.7a | 3.9a | 11.3a | 14.8a | 10.3a |
|  | 3d | 0.9b | 4.9b | 8.7b | 18.6a | 3.4a | 9.4b | 14.5 | 10.3 |

Table 1. Responses of height and leaf number among different EMS and SA treatment time.

| Mutagen | Rate | h1 | h2 | n3 | n4 | 11 | 12 | 13 | 14 |
| :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: | :---: |
| EMS | 0 | 0.9b | 4.7 a | 8 8 | 9 a | 4ab | 9.9a | 15.2a | 17.6a |
|  | 30 | 0.7 bc | 5.4a | 7.9a | 10.1a | 3.8ab | 10.1a | 14.3a | 19.5a |
|  | 60 | 0.5bc | 4 a | 6.9a | 8.3a | 2.5bc | 9.9a | 13.6a | 17.8a |
|  | 90 | 0.2b | 3.2a | 7.8a | 11a | 1.7 c | 9.8a | 13.3a | 18.5a |
|  | 120 | 1.6a | 3.5a | 9.3 a | 7.3a | 5a | 7a | 17a | 16a |
|  | 150 |  |  |  |  |  |  |  |  |
| SA | 0 | 1 a | 5.6ab | 9.9 a | 12a | 3.7ab | 10.1a | 15.1a | 20.1a |
|  | 2 | 1.1a | 501b | 7.9a | 8.7 b | 3.3ab | 10.5a | 14a | 18.3a |
|  | 4 | 1 a | 5.9ab | 9.1a | 10.8ab | 4.3a | 11.1a | 15.1a | 18.8a |
|  | 6 | 1.2a | 6.6a | 10.5a | 11ab | 3.5ab | 10.6a | 15.4a | 18.7 a |
|  |  | 1.2a | 7 a | 10.6a | 11.3ab | 3.1b | 11.6a | 15.8a | 18.9a |
|  | 10 | 1.3a | 6.9a | 10.4a | 11.6ab | 3.4ab | 10.3a | 14.9a | 20.8a |

Table 2. Responses of height and leaf number among different EMS and SA treatment solution.


Figure 3. Responses of abnormal leaf number among EMS and SA treatment solution.


Figure 4. The abnormal bud and abnormal leaves.


Figure 5. The standard of rating.

## Results

$>$ The median lethal does(LD50) is about 60-90 mM for EMS and 8-10 mM for SA.
$>$ Plants treated with 60, 90 mM EMS or $\mathbf{8 - 1 0} \mathbf{m M}$ SA have the medium visual damage rating score. This rating result further confirm the LD50 conclution.
> The average of height and leaf number between LD50 condition and control are similar. The anova test shows no significant difference,this may be caused by the mutagen enlarge the ingroup variation.
> As the abnormal leaf number of SA decreased from $6.3(2 \mathrm{mM})$ to $1(8 \mathrm{mM})$, EMS increased from $5(\mathrm{mM})$ to $7.8(120 \mathrm{mM})$
> There was no significant difference of chlorophyll content, branch number among all EMS and SA treatment solution(data not shown).

## Conclusion

$>$ Plants can be tested with EMS at rate up to 90 mM for 2d, and with SA at rate up to 10 mM for 3d as the optimum doses of chemical mutagens.
> This is just the preliminary experiment results, after flowering, the folwer number, folwer size, flower color datas will help further optimize the doses of EMS and SA mutagens.

