



**GUIDELINES FOR APPLICATIONS ON  
GENETICALLY MODIFIED ORGANISMS  
WITH STACKED GENES IN GHANA**

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## DECLARATION

I, **Prof. Charles ANTWI-BOASIAKO**, the Chairman of the Board of the National Biosafety Authority (NBA), acting under Section 40(3) of the Biosafety Act, 2011 (Act 831), hereby issue these Guidelines.

Dated this 30th day of October 2023.



**Prof. Charles ANTWI-BOASIAKO, PhD**

## 1.0 INTRODUCTION

The combination of two or more genes of interest into an organism is known as gene stacking. Stacked traits are the combined traits that emerge from this approach. Gene stacking can be accomplished using molecular technology or conventional breeding. Traits from two or more genetically modified (GM) events can be combined using conventional breeding methods. These are commonly referred to as "Breeding Stacks." The term "Molecular Stacks" is used to describe instances where two or more new genes have been stacked using molecular methods.

Crops with stacked genes could present numerous advantages such as increased productivity, reduced use of chemicals, cultivation of plants in adverse environmental conditions, and higher nutritional quality. For instance, they have offered enhanced seeds that combine genes for herbicide tolerance and insect resistance. Since their introduction, they have been appreciated by many researchers and farmers for their adaptability, improved traits, and low cost, and are currently driving the development of Genetically Modified Organisms (GMOs). These advantages are leading to an increase in their adoption in many parts of the world.

The National Biosafety Authority (NBA) is mandated to ensure that gene stacking does not create unintended effects and changes that require additional safety assessments. The Biosafety Act, 2011 (Act 831) defines GMOs to include any organism that has been transformed by the insertion of one or more genes, or regulatory elements, or an organism that has had its own genes modified without the insertion of any new genes and their products. Molecular Stacks and Breeding Stacks are GMOs that under the Biosafety Act, 2011 (Act 831) require risk assessment and risk management. However, unlike Molecular Stacks which will result in new events, GM-stacked products produced via conventional breeding do not result in a new event(s) and therefore do not require risk assessment and risk management. In other words, the individual events that are stacked through conventional breeding typically would have undergone regulatory assessments by regulators either locally or globally. Thus, in consideration of the processes leading to the development of these stacks, biosafety regulations for risk assessment will be applied on a case-by-case basis. Considering that Molecular Stacks result in new events that have to undergo the regulatory process as a GMO, these Guidelines will emphasize possible scenarios involving GM Breeding Stacks.

## 2.0 SCOPE

These Guidelines shall apply to any person(s) or institution(s) in Ghana intending to introduce crops with stacked traits into the environment for cultivation or for use as food, feed, and for processing.

These Guidelines further emphasize possible scenarios involving GM Breeding Stacks where:

- a) the single events have already undergone regulatory approvals in Ghana or globally.
- b) one or more of the single events have not undergone regulatory approvals in Ghana or globally.
- c) none of the single events has undergone regulatory approvals in Ghana or globally.

## 3.0 OBJECTIVE

To provide information and guidance to potential applicants on the requirements and procedures for the regulation of GM stacked events developed through conventional breeding ("Breeding Stacks") or molecular techniques ("Molecular Stacks").

## **4.0 DETERMINATION OF THE RISK ASSESSMENT PROCESS FOR GMOs WITH STACKED EVENTS**

### **4.1 BREEDING STACKS**

#### **A. Stacking of GM Events through conventional breeding where the single events have already undergone regulatory approvals in Ghana or globally.**

- (1) An applicant under this provision shall make a notification to the NBA with all relevant information for review and decision making. The notification shall entail:
  - a. A confirmation of the intactness and stability of inserted genetic elements;
  - b. An assessment of potential interactions between combined events and the resulting phenotypic traits in the case that the potential for interactions cannot be excluded based on the individual modes of action of the respective GM Events;
  - c. In case the potential for interactions cannot be excluded based on the individual modes of action of the respective GM event(s) (see point (b) above), an assessment of potential interactions of the stacked events on the conservation and sustainable use of biological diversity in the likely potential receiving environment, also taking into account risks to human health, depending on the scope of the application.
- (2) Approval of the higher order stacks covers the intermediate stacks and single events previously authorized.
- (3) History of safe use of single events / List of approvals issued by other countries
- (4) Other additional and key considerations for various categories of breeding stacks are summarized in Table 1 provided in Annex I of these Guidelines.

#### **B. Stacking of GM Events through conventional breeding where one or more of the single events have not undergone regulatory approvals in Ghana or globally.**

- (1) The applicant shall submit a complete application for the unapproved events and a notification for the higher order stacks.
- (2) A simultaneous review process will be adopted for the unapproved single event and the higher order stack (final stacked product) to aid in decision making.

For higher order stack, the applicant will be required to provide:

- i. A confirmation of gene expression levels of each of the single events
  - ii. An assessment for possible interactions – synergistic or antagonistic
  - iii. A confirmation of event intactness and stability
  - iv. History of safe use of the single events / List of approvals issued by other countries.
- (3) The approval of the higher order stack will also cover the intermediates and their singles, where applicable.
- (4) Other additional and key considerations for various categories of Breeding Stacks are summarized in Table 1 provided in Annex I of these Guidelines.

### **C. Stacking of GM Events through conventional breeding where none of the single events has undergone regulatory approvals in Ghana or globally.**

- (1) The applicant shall submit a complete application for each single event and a notification for the higher order stack.
- (2) A simultaneous review process may be adopted for the single events and the higher order stack to ensure efficient decision making.
  - For higher order stack, the applicant will be required to provide:
    - i. A confirmation of gene expression levels of each of the single events
    - ii. An assessment for possible interactions – synergistic or antagonistic
    - iii. A confirmation of event intactness and stability
    - iv. History of safe use of the single events / List of approvals issued by other countries.
- (3) The approval of the higher order stack will also cover the intermediates.
- (4) Other additional and key considerations for various categories of Breeding Stacks are summarized in Table 1 provided in Annex I of these Guidelines.

## **4.2 MOLECULAR STACKS**

Stacked events development using molecular techniques could be carried out through co-transformation, re-transformation, or the use of multi-gene cassettes. Regardless of the method used, the following parameters will be assessed prior to release:

### **(a) Assessment of the Intactness of the Inserted Loci and Genotypic Stability**

The following should be provided by the applicant:

- (i) Information on the genetic stability of the transgenic locus (loci); phenotypic stability and inheritance pattern of the introduced genes
- (ii) Information showing that the integrity of the genes is retained in the GMO.
- (iii) Source of the genetic material, the sampling design, and the number of plants used for the analysis. When analyzing the inheritance pattern, appropriate statistical methods should be applied, and
- (iv) A comparison between the structure of the inserts in stacked traits and that of the insert present in the corresponding event(s).

### **(b) Assessment of Potential Interactions**

Whenever two or more traits are stacked together in one crop, the potential for these traits to interact and any safety concerns these may raise, need to be evaluated. Applicants will therefore be required to provide information on the function of the nucleic acid region intended for insertion. This information should comprise the following elements:

- (i) complete sequence of the nucleic acid intended to be inserted, including information on the donor organism and any deliberate alteration to the corresponding sequence in the donor organism,
- (ii) history of safe use of the gene product arising from the regions intended for insertion, and
- (iii) data on the possible relationship of the gene product with known toxins, anti-nutrients,

and allergens.

**(c) Assessment of Effects of GM Stacked Events on the Conservation and Sustainable Use of Biological Diversity**

For instances of intended introduction into the environment, the objective of the analysis is to assess the potential impact of the stacked events on the conservation and sustainable use of biological diversity in the likely potential receiving environment. The applicant is therefore required to provide information on the phenotypic characteristics, including the modified traits, compared to the conventional counterparts.

**(d) Compositional Assessment Using Substantial Equivalence**

The analysis of the nutritional composition of the crop shall be assessed relative to its conventional counterpart (substantial equivalence). The relevant food safety assessment guidelines (e.g., CODEX) shall apply.

## **5.0 GM STACKS IMPORTED FOR USE AS FOOD, FEED, OR FOR PROCESSING**

GM stacked events imported as part of a consignment for direct use as food, feed, or for processing (and not for cultivation) would be regulated in accordance with NBA's existing procedures for the Importation of Genetically Modified Organisms for Food, Feed, or for Processing.

## **6.0 DECISION MAKING ON GM EVENTS STACKED THROUGH CONVENTIONAL BREEDING**

Once the single events and higher order stacks are approved by the NBA, any other sub-combinations of those events, with the exception of sub-combinations that develop through natural segregation in cultivated fields of approved higher order stacks, will require a request to the NBA for approval for use as parental lines or for commercialization.

## **7.0 POST RELEASE MONITORING PLAN**

### **General Surveillance**

1. Applicants are required to put in place a plan for general surveillance after the release period.
2. Applicants may adopt different approaches for surveillance which could include but not limited to:
  - (a) monitoring of stacked events, the process, and their cultivation sites by using appropriate survey instruments as determined by the NBA; or
  - (b) farmer surveillance and complaint systems.
3. Applicants should carry out post-harvest monitoring and evaluation of any risk.

## **8.0 DEFINITION OF TERMS**

For the purpose of these Guidelines:

“**Act**” the Biosafety Act, 2011 (Act 831).

“**Applicant**” is any person who submits an application pursuant to a provision of the Biosafety Act.

“**Breeding Stacks**” is a GMO containing combined traits from two or more GMOs resulting from conventional breeding techniques.

“**Conventional Breeding**” involves identifying parent plants with desirable characteristics to create favorable combinations in the next generation.

“**Co-transformation**” transformation with two or more independent new genes.

“**DNA**” means deoxyribonucleic acid.

“**Event**” is a genotype produced from the transformation of a species using a specific genetic construct.

“**Gene**” is a distinct sequence of nucleotides forming part of a chromosome, the order of which determines the order of monomers in a polypeptide or nucleic acid molecule that a cell (or virus) may synthesize.

“**Gene Pyramiding**” multiple genes controlling a single trait.

“**Gene Stacking**” is the combination of two or more genes of interest in the genome of a single plant.

“**Higher Order Stack**” refers to the desired final product containing the highest combination of single events that will eventually go to the end users.

“**Intermediates**” is a combination of two or more GM events within a higher order stack. Has the same meaning as sub-stacks.

“**Molecular Stacks**” is a plant transformed by using molecular methods, where two or more traits are simultaneously (Co-Transformation) or sequentially (Re- Re-transformation) introduced into a host plant by standard delivery systems such as Agrobacterium-mediated and biolistic methods.

“**Notification**” is a brief letter outlining the combined traits and stating that the stack expresses these traits in a manner similar to the parental lines.

“**Parental line**” is a plant of a desirable trait that is crossed with another. These are inbred lines that are crossed with each other for purposes of seed production.

“**Stacked Events**” is the creation of a genetically modified organism (GMO) with more than one genetic modification. This can be done by (a) cross-breeding two GMOs with each having one or more pre-existing modifications (b) carrying out a second genetic modification in an existing GMO or (c) introducing multiple genes or traits at once.

“**Synergistic Effect**” is an interaction of elements that when combined produce a total effect that is greater than the sum of the effect of the individual elements.

“**Transformation**” is the specific process where exogenous genetic material is directly taken up and incorporated by a cell through its cell membrane.

“**Re-transformation**” is the use of modern biotechnology to produce a GMO where the recipient plant is already a GMO i.e., a plant harboring a transgene is transformed with other transgenes.



## 9.0 ANNEX I: KEY CONSIDERATIONS FOR VARIOUS CATEGORIES OF BREEDING STACKS

**Table 1: Key Considerations for Various Categories of Breeding Stacks**

Category	Considerations	
A	Stacking of GM Events through conventional breeding where the single events have been approved.	<p>Applicant to submit a notification to the NBA with information including:</p> <ul style="list-style-type: none"> <li>• Confirmation of gene expression levels of each of the single events</li> <li>• An assessment for all possible interactions-synergistic or antagonistic</li> <li>• Confirmation of event intactness and stability</li> <li>• Approval of the higher order stacks covers the intermediate stacks and single events previously authorized.</li> <li>• History of safe use of single events / List of approvals issued by other countries</li> </ul>
B	Stacking of GM Events through conventional breeding where one or more of the single events have not been approved.	<p>Applicant to submit a complete application for the unapproved event and a notification for the higher order stack.</p> <ul style="list-style-type: none"> <li>• A simultaneous review process will be adopted for the unapproved single events and the higher order stack (final stacked product). For higher order stack, the applicant will be required to provide: <ul style="list-style-type: none"> <li>i. A confirmation of gene expression levels of each of the single events</li> <li>ii. An assessment for possible interactions – synergistic or antagonistic</li> <li>iii. A confirmation of event intactness and stability</li> <li>iv. History of safe use of the single events / List of approvals issued by other countries</li> </ul> </li> <li>• The approval of the higher order stack will also cover the intermediates and their singles, where applicable.</li> </ul> <p><b>NB:</b> CFT data will only be required for the combination product(s) that the applicant intends to make commercially available to end users and in accordance with existing Guidelines. Where the</p>

		<p>applicant intends to commercialize any sub-combination other than the ones approved, supplemental data may be requested, as appropriate. Where possible, modalities for supplemental data transportability or bridging may be explored.</p>
C	<p>Stacking of GM Events through conventional breeding where none of the single events have been approved.</p>	<ul style="list-style-type: none"> <li>• Applicant to submit separate complete applications for all unapproved single events and a notification for the higher order stack.</li> <li>• A simultaneous review process will be adopted for all the unapproved single events and the higher order stack (final stacked product). For higher order stack, the applicant will be required to provide:             <ol style="list-style-type: none"> <li>i. A confirmation of gene expression levels of each of the single events</li> <li>ii. An assessment for possible interactions – synergistic or antagonistic</li> <li>iii. A confirmation of event intactness and stability</li> <li>iv. History of safe use of the single events / List of approvals issued by other countries.</li> </ol> </li> <li>• The approval of the higher order stack will also cover the intermediates.</li> </ul> <p><b>NB:</b> CFT data will only be required for the combination product(s) that the applicant intends to make commercially available to end users and in accordance with existing Guidelines. Where possible, modalities for supplemental data transportability or bridging may be explored.</p>
<p><b>NB:</b></p> <ul style="list-style-type: none"> <li>• In all cases, once the single events in higher order stacks are approved by the NBA, any other intermediate event will require a notification to the NBA for approval for use as a parental line or for commercialization.</li> <li>• Natural segregation leading to possible stacking of genes in cultivated fields will not lead to a notification to the NBA.</li> </ul>		

**10.0 ANNEX 2: STACKED EVENT FLOW CHART**

