better late than never...

29th International Seminar on Interaction of Neutrons with Nuclei (ISINN-29)

# OBTAINING OF INITIAL FORMS FOR SYNTHETIC SELECTION OF DROUGHT-RESISTANT RICE CROPS USING RADIATION MUTAGENESIS ON FAST NEUTRONS

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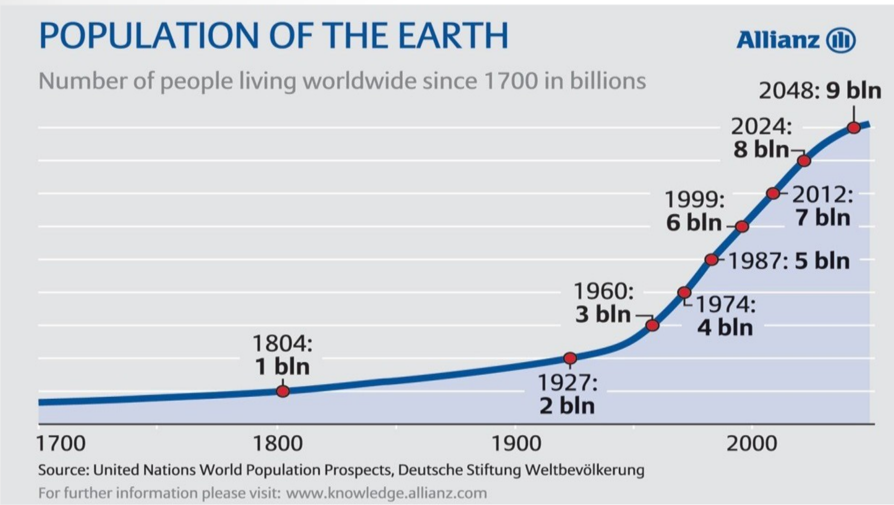
**Frank Laboratory of Neutron Physics, JINR, Dubna, May 29th - June 2nd, 2023**

# Induced mutation breeding

*First used in 1928* by Stadler, increases the frequency of mutations to several orders of magnitude compared to natural spontaneous mutation rates, as a result it provides diverse variability to breeders to select and improve upon




Ionizing radiation

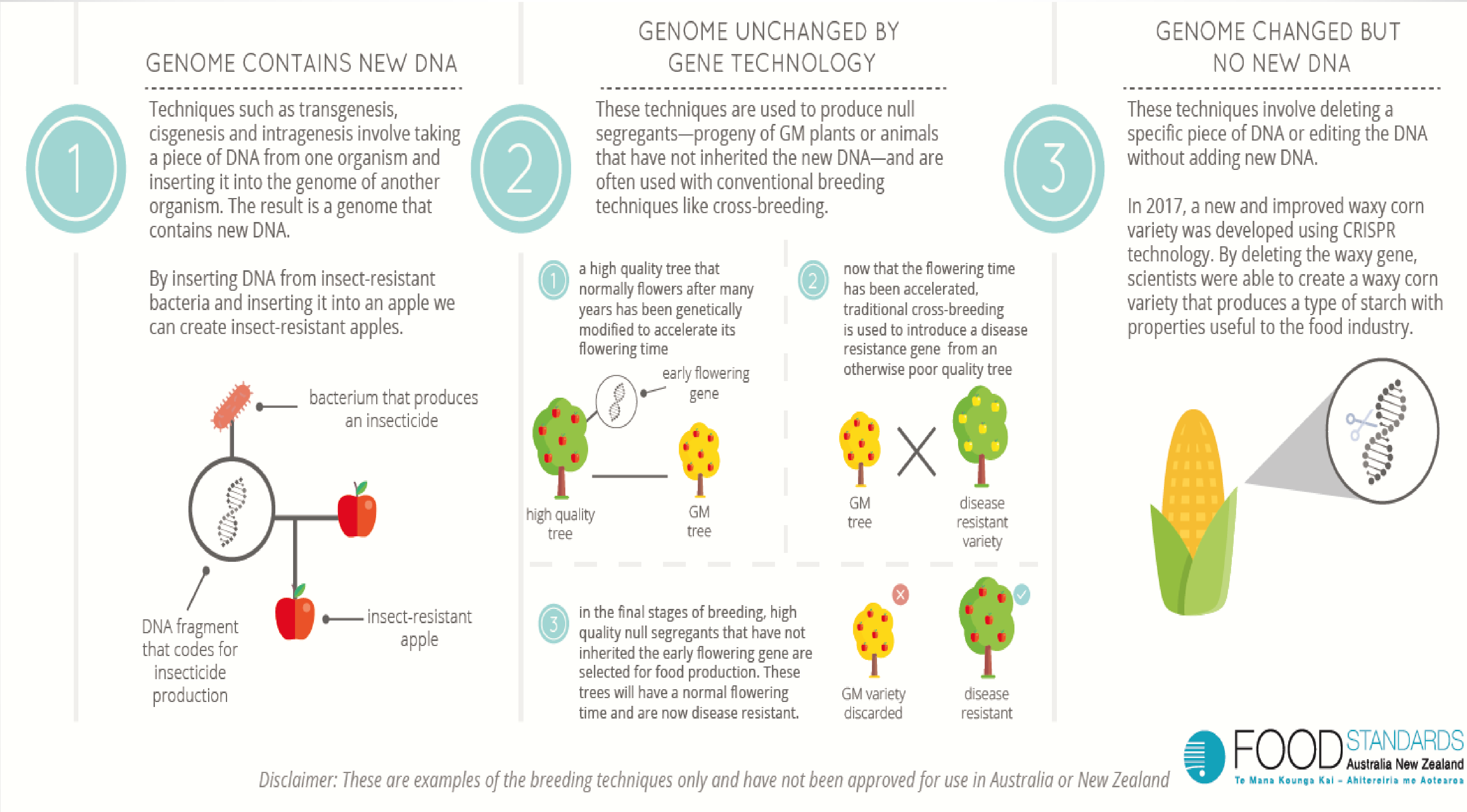
Wing R.A. et. al. The rice genome revolution: from an ancient grain to Green Super Rice // Nat Rev Genet. 2018.

# Yield losses in some major crops caused by drought

| Crop species | Stress | Yield losses (%) | Reference |
|---|---|---|---|
| Maize (*Zea mays* L.) | Drought | 63–87 | Kamara et al., 2003 |
| | Heat | 42 | Badu-Apraku et al., 1983 |
| Wheat (*Triticum aestivum* L.) | Drought | 57 | Balla et al., 2011 |
| | Heat | 31 | Balla et al., 2011 |
| Rice (*Oryza sativa* L.) | Drought | 53–92 | Lafitte et al., 2007 |
| | Heat | 50 | Li et al., 2010 |
| Chickpea (*Cicer arietinum* L.) | Drought | 45–69 | Nayyar et al., 2006 |
| Soybean (*Glycine max* L.) | Drought | 46–71 | Samarah et al., 2006 |
| Sunflower (*Helianthus annuus* L.) | Drought | 60 | Mazahery-Laghab et al., 2003 |

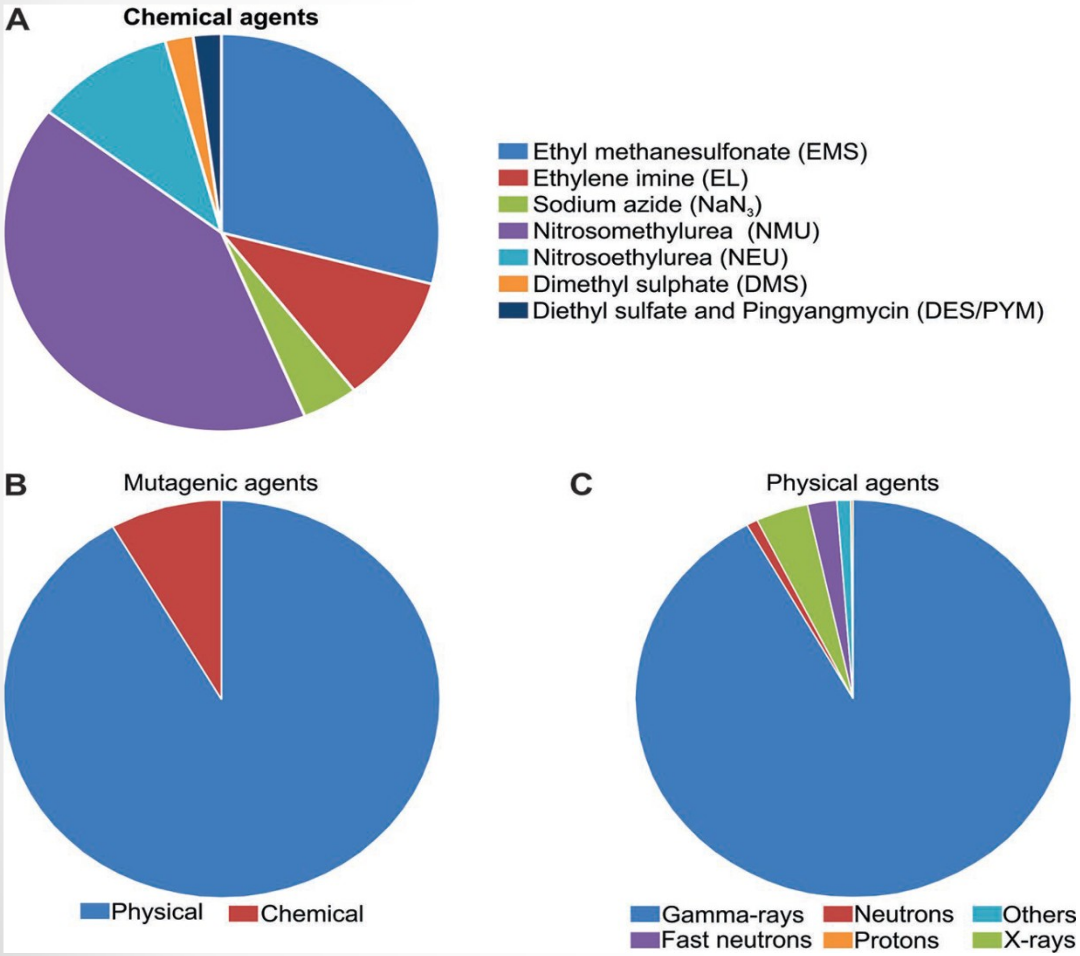
Fahad, S., Bajwa, A. A., Nazir, U., Anjum, S. A., Farooq, A., Zohaib, A., … Huang, J. (2017). Crop Production under Drought and Heat Stress: Plant Responses and Management Options. Frontiers in Plant Science, 8. doi:10.3389/fpls.2017.01147

# New Plant Breeding Techniques (NBTs)




https://www.foodstandards.gov.au/Pages/default.aspx.2021

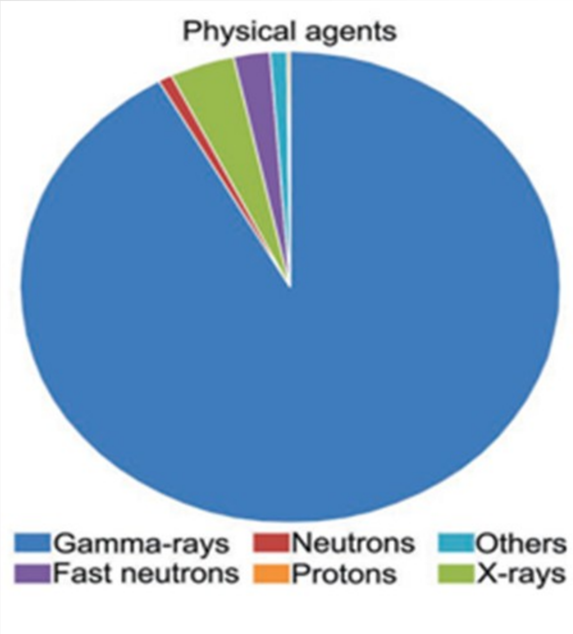
# Mutagens applied in rice mutagenesis according to FAO/IAEA-MVD




**(A) Chemical mutagens;**
**(B) Chemical and physical agents;**
**(C) Physical mutagens.**

V.E. Viana, C. Pegoraro, C. Busanello and A. Costa de Oliveira (2019). Mutagenesis in Rice: The Basis for Breeding a New Super Plant. Front. Plant Sci. 10:1326. doi: 10.3389/fpls.2019.01326

# Fast neutrons as a type of radiation mutagenesis



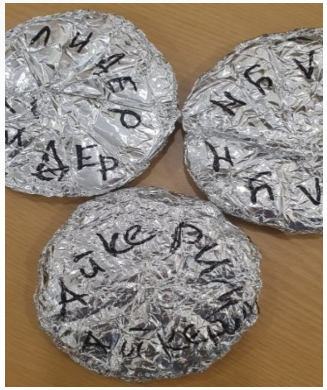

*Physical mutagens applied in rice mutagenesis according to FAO/IAEA-MVD*

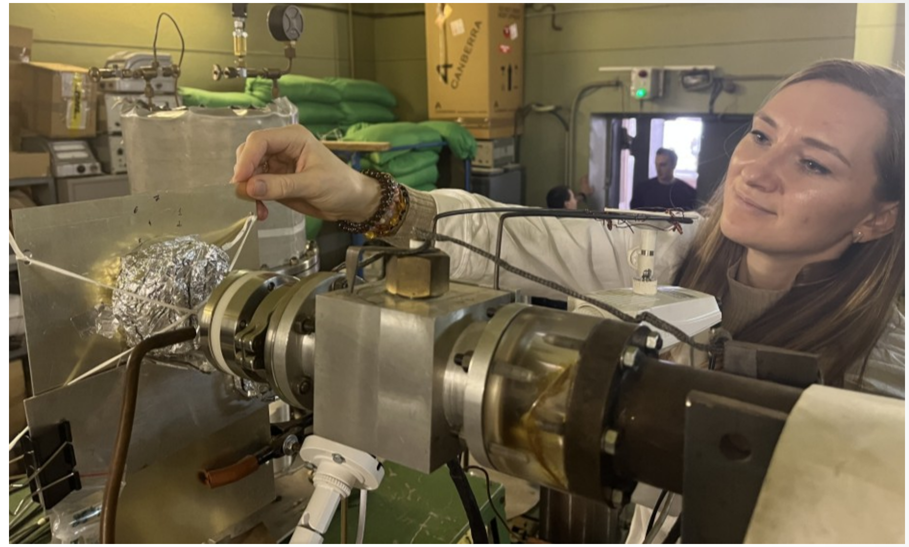
- higher energy compared to $\gamma$ rays, especially in biological materials (*H*- high interaction cross-section with *n*)
- neutron capture and elastic scattering with C, H, O, N nuclei
- generation of direct and indirect ionizing radiations

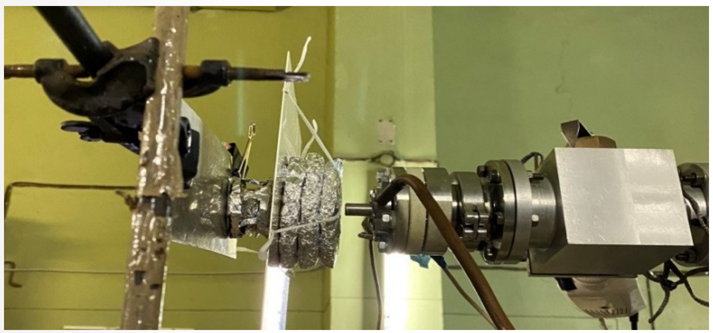
- **Increase the frequency of chlorophyll mutations**
- **Changes in amylose content not alter the amylopectin structure**
- **Changes in the *FRO1* gene → tolerance to iron toxicity**
- **Deletions, inversions, translocations, tandem duplications**
- ***NPR1 homologue* → immune responses against bacterial infection by *Xanthomonas oryzae***


Kadam, S.T., Vishwakarma, G., Kashyap, Y. et al. Thermal neutron as a potential mutagen for induced plant mutation breeding: radiosensitivity response on wheat and rice. Genet Resour Crop Evol 70, 789–798 (2023). https://doi.org/10.1007/s10722-022-01461-z

# Fast neutron irradiation







- $D(d,n)\,^3He$
- $E_n = 4.1$ MeV
- $I \sim 1,7 - 2\mu A$
- $E_D = 2.5$ MeV
- neutron monitor of the PIXE-4 type
- 204 hours
- 35 - 40 million particles /hour

*Experimental site LLP "Kazakh Research Institute of Rice named after. I. Zhakhaev" in the Republic of Kazakhstan.*



*The study performs in the scope of the Ministry of Agriculture of the Republic of Kazakhstan project number BR10765056*

# Results of fast neutron irradiation

*Mutant samples where green rice grains do not contain rice seeds*

*Mutant samples that shows lag the maturation period (top is the Syr-Syluy control variety, bottom is irradiated samples).*

*Aikerim*

*Leader*

*Syr-Suluy*

*Photo of the control plants (on the left) and M1 plants distinguished by resistance to salinity and drought.*

- received **mutant forms** resistant to salinity (NaCl) and drought factors (sorbitol). The largest number of mutant forms was obtained from variety Syr Suluy, followed by Leader and Aikerim varieties
- it was discovered that the median lethal dose (LD50) for fast neutrons – 50 Gy
- M1 plants differ significantly from the original forms in **morphological features** : plant height, length, stunting and dwarfism (40-80 cm), and high empty-grain of panicles (up to 100%)

# Results of fast neutron irradiation

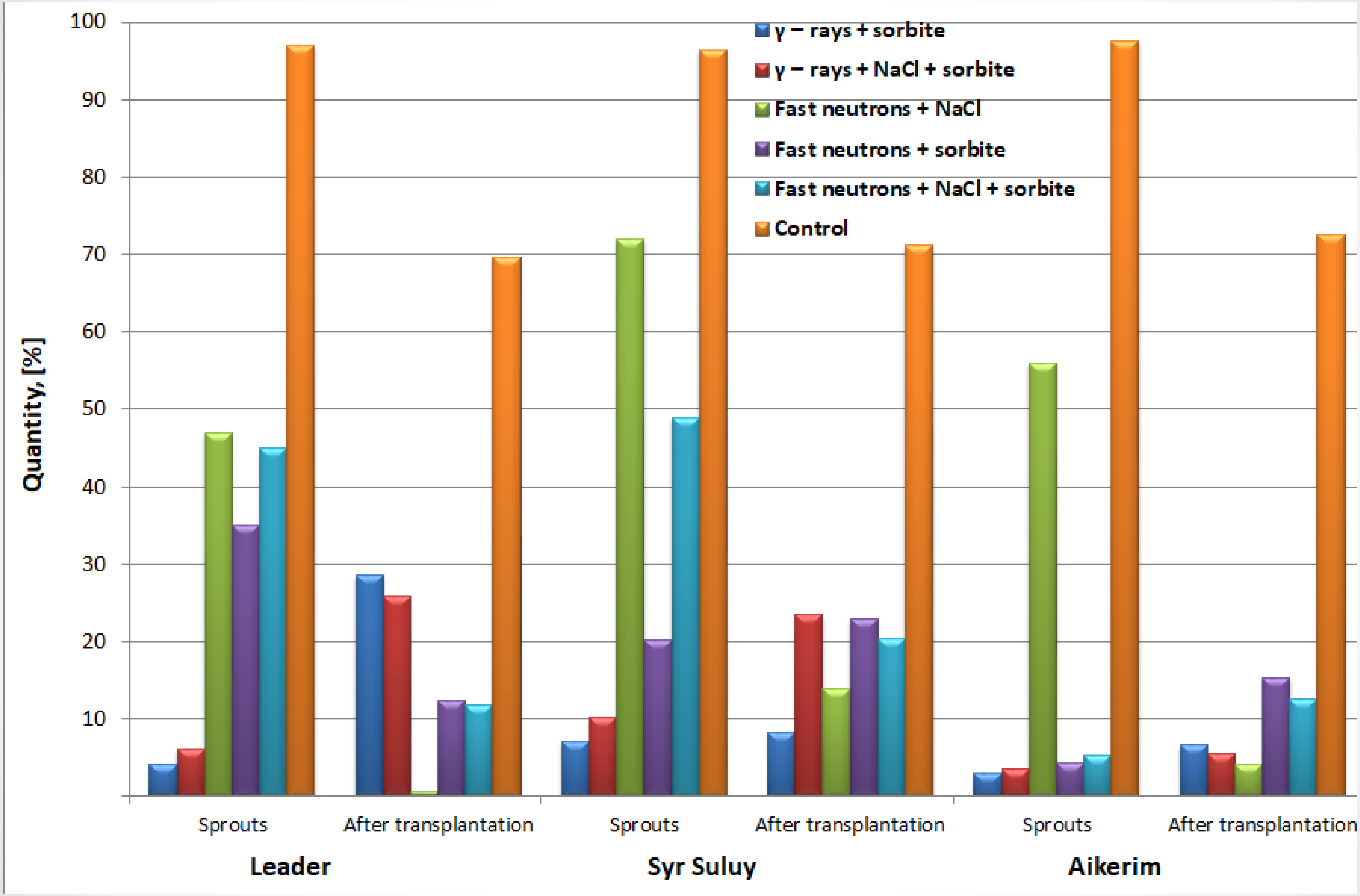



*Figure 1. Laboratory germination and survival of seeds treated with γ-rays, fast neutrons and salinity (NaCl) and drought factors (sorbitol)*

# Conclusion

- ❖ It was discovered that the median lethal dose (LD50) for fast neutrons – 50 Gy
- ❖ Received mutant forms (max Syr Suluy), resistant to the stressful conditions of the Kazakhstan region of the Aral Sea (salinity, sorbitol)
- ❖ Mutant forms are going to be used as initial forms in synthetic selection
- ❖ A distinct dependence on the effect of ionizing radiation and stress factors on the number of induced resistant mutant forms has been related to the initial rice variety.

**Acknowledgments:**
The authors wish to acknowledge Belarus-JINR Program № 308 items №21,22; RO-JINR Program No. 366/2021 item 84, Serbia-JINR Program No. 373/2023 items 4 and 5, Vietnam-the International Center of Physics at the Institute of Physics Grant ICP.2022.04.

# Th u for attention!