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TITLE

Pollen mitochondria in cytoplasmically male
sterile tobacco zygotic and embryonic cells

FINAL REPORT FOR THE PERIOD

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F I N A L R E P O R T

Your ref: 302-D₂-GRE-3020-1

- 1.a. (I) Contract Number : 3020/R1/RB
 (II) Title of the project : Pollen mitochondria in cytoplasmically male sterile tobacco zygotic and embryonic cells.
 (III) Institute where research is being carried out: Athens Agricultural College.
 (IV) Chief Scientific Investigator: Dr.Ing.Y.N.Symillides
 (V) Time period covered 1/5/1984 - 31/4/1985.

b. Description of the research carried out:

The main objective of our research is to follow the fate of paternal cytoplasmic organelles i.e. mitochondria and plastids. New series of specimens were prepared and examined with both light and electron microscopes. Tobacco inflorescences with five, 0,6 - 1cm long, flower buds, were placed in solid nutrient media of the following consistency:

	mgr/lt		mgr/lt
A: NH ₄ NO ₃	1,650.000	B: KI.	0.821
KNO ₃	1,900.000	CuSO ₄ 5H ₂ O	0.025
CaCl ₂ 2H ₂ O	440.000	Na ₂ MoO ₄ 2H ₂ O	0.250
MgSO ₄	181.000	CoCl ₂ 6H ₂ O	0.025
KH ₂ PO ₄	170.000		
MnSO ₄ H ₂ O	16.900		mgr/lt
ZnSO ₄ 7H ₂ O	8.600	C: FeSO ₄ 7H ₂ O	24.900
H ₃ BO ₃	6.200		

Solid nutrient medium was then prepared in proportions A, B, C : 200: 1 : 1 with 1 per cent of agar. pH was adjusted to 6.2 and then tritiated thymidine was added at 10 μ Ci/flower bud.

Pollen collected from the flowers which had come to anthesis was used for artificial pollination of non treated stigmas. The fertilized eggs were then processed for autoradiography.

- c. Results: Part of the results obtained mainly by the use of leucine ¹⁴C will be presented at FAO-IAEA International symposium (see enclosed summary).
Results obtained employing tritiated thymidine are as follows:
1. Pollen grains prelabelled with radioactive precursor present less silver grains fixed than pollen from leucine ¹⁴C experiments (Fig 1a and b).
 2. Radioactivity was detected in zygotic cells (Fig. 2) as well as in young proembryos (Fig. 3 a, b)
 3. No apparent labelling was observed in the plastids or mitochondria of the so far examined material.

Conclusions: As we have also mentioned in the enclose summary, radioactive precursor does not impair pollen viability and fertility, therefore, the process of fertilization may be followed.

Few tagged organelles observed in the embryo sac, mainly mitochondria, may have some direct or indirect relation with the paternally originated cytoplasm. The latter hypothesis is also supported by the thymidine ³H experiments, where no labelled organelles have been so far observed.

Further examination of the material is considered to be necessary since the labelling of pollen cytoplasm with thymidine ³H is relatively poor and consequently the tracer in embryo sac is very sparse.



Dr. Ing. Y. N. Symillides.

(chief scientific investigator)