

complement of the Wild Pig from Japan (*Sus vittatus leucomystax*), according to the findings of MURAMOTO et al.⁹

Since at least one of our samples came from the area in which a chance for a cross with domestic breed never existed (Prokletije – the area inhabited by Moslems who for religious reasons never raised pigs), the differences between chromosome complements of Balkan Wild Pig populations and those from Germany suggest a case of intraspecific chromosomal polymorphism with at least 2 chromosomal types ($2n = 36$ and $2n = 38$). There is little doubt that Domestic Pig originated from the 38-chromosomes type which inhabits southern and eastern parts of Europe and probably most of Asia. Further studies are necessary to find out the exact areas of the 36- and 38-chromosomes types and to shed more light on the nature of the Robertsonian changes which caused this variation¹⁰.

Résumé. Le complément chromosomique des sangliers des régions de l'Est et du Sud de la Yougoslavie est caractérisé par un nombre diploïde $2n = 38$ et il est composé de 6 paires d'autosomes acrocentriques et de 12 paires d'autosomes méta- et submétacentriques, ce qui signifie qu'il est identique au complément du porc domestique et qu'il diffère du complément chromoso-

mique des sangliers d'Allemagne. Il est évident qu'il s'agit là d'un cas de polymorphisme chromosomique intra-spécifique avec au moins deux types de nombre diploïde ($2n = 36$ et $2n = 38$) résultant des variations de Robertson. Il est très probable que le porc domestique tire son origine d'un type à 38 chromosomes.

S. ŽIVKOVIĆ, V. JOVANOVIĆ,
I. ISAKOVIĆ and M. MILOŠEVIĆ

*Institute for Biological Research, Faculty of Science,
and Institute 'Dr. Ilija Djuričić', Belgrade (Yugoslavia),
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¹⁰ We thank to the managers of 'Jelen' and 'Morović' Game Preserves through whose kind cooperation the studied animals were provided.

Autoradiographic Evidence for a Thymidine Precursor Pool in the Root of *Allium cepa*

In order to determine the possible presence of a thymidine precursor pool in the onion root (*Allium cepa*), the regional pattern of DNA synthesis was followed by means of a tritiated-thymidine (³H-TdR) continuous labeling procedure coupled with autoradiography.

Methods. A commercial onion bulb (*Allium cepa* L., $2n = 16$) was grown in a vial of tap water at room temperature until the growing roots reached a length of approximately 2 cm. The bulb was then transferred to a solution of ³H-TdR with a concentration of 5 μ C/ml (specific activity 6.25 C/mM), and representative root were removed and fixed in Carnoy's fixative following a 24 h continuous label in ³H-TdR and after 1, 8, and 20 h chase periods in unlabeled tap water.

The roots were embedded and sectioned longitudinally at a thickness of 8 μ m and stained by the Feulgen method¹. All the autoradiograms were prepared by the stripping-film method², incubated at 4°C in the dark for 10 days, and developed together.

The percentage of labeled nuclei was determined in the meristem and in the elongation region. Nuclear grain counts were determined in autoradiographs for each chase interval in the respective regions with the aid of a quadrille-reticle having 400 squares. Grain counts were corrected for background by counting, in each autoradiograph nearest each of the regions under study, the number of grains in a known area of film with no underlying root tissue.

Results. The percentage of nuclei showing incorporation of ³H-TdR in the meristem and elongation region is shown in Table I. Note that for every chase period, greater percentages of nuclei are labeled in the elongation region as compared to the meristematic region. It is of particular interest that in the meristematic region there is a pronounced elevation in the percentage labeled nuclei preceding a progressive decline. It can be seen

from Table II, that for each chase interval the average nuclear grain count of the elongation region is significantly higher than that of the meristematic region. Furthermore, with increasing chase periods the nuclei of both regions show a progressive increase in grain counts.

Discussion. One might predict a continual decrease in the percentage labeled nuclei in the meristematic region due to division and displacement of nuclei by the growing meristem. The initial rise in percent labeled meristematic nuclei (Table I) is interpreted as evidence for the presence

Table I. The percent of nuclei labeled after exposure to ³H-TdR for 24 h and subsequent exposures to unlabeled medium

Amount of time in unlabeled medium (h)	Nuclei labeled (%)	
	Meristem	Elongation
0	75 ^a	87
1	90	99
8	82	86
20	69	85

A minimum of 6 roots were utilized for each interval. The 0 h sections correspond to roots exposed to ³H-TdR for 24 h and sacrificed immediately after. The 1, 8, and 20 h sections correspond to roots subsequently exposed to unlabeled medium for 1, 8, and 20 h. ^a

¹ R. D. LILLIE, *Histopathologic Technic and Practical Histochemistry* (McGraw-Hill Book Co., New York 1965), p. 149.

² G. E. STONE and D. M. PRESCOTT, *J. Cell Biol.* 21, 275 (1964).

of a ^3H -TdR precursor pool. This pool effect is reflected by a parallel pattern in the percentage of labeled nuclei in the elongation region during the chase. It was also observed by SAMPSON and DAVIES³ in *Vicia faba*, that the elongation region contains more heavily labeled nuclei than does the meristematic region. This is in agreement with the studies of PELC and LA COUR⁴, who found intense labeling in approximately 25% of the cells at a distance of 3–5 mm behind the meristem. SAMPSON and DAVIES³, also found that after a 4 h label of ^3H -TdR followed by a 48 h chase treatment, the amount of incorporated isotope increased even after the external source of isotope was removed. This could be due to a possible use of a precursor pool of ^3H -TdR in the cells of both regions formed during the preceding S period. In support of this idea, it was observed that in *Tetrahymena pyriformis* a pool of TdR derivatives is formed only during the S phase, and when DNA synthesis is completed, remains in the nucleus until the next round of DNA synthesis occurs after cell division^{2,5}. This was also confirmed in the grasshopper neuroblast⁶.

DNA synthesis also occurs in the elongation region although there are no mitotic figures observed, and consequently, there is no grain count dilution. The data in Table II, show an increase in grain counts in the nuclei of the elongation region, and hence an increase in DNA synthesis during the chase period. This suggests the

existence of a ^3H -TdR precursor pool in the cells of the elongation region. This finding is in agreement with the autoradiographic data of WOODARD et al.⁷ on the roots of *Vicia faba*, who also found this grain count elevation in the elongation region during the chase period after pulsing with ^3H -TdR. They also observed in this root a large ^3H -TdR precursor pool in the elongation region.

In accordance with the aforementioned authors^{2,5-7}, the conclusion seems justified that there is a large ^3H -TdR precursor pool in the elongation region as seen by the higher grain counts in this region (Table II).

A similar pool of phosphorylated derivatives of ^3H -TdR has been detected by means of autoradiographic methods in cells of the mouse bone marrow⁸, in *Tetrahymena*⁹ and, in mouse cells in culture^{10,11}.

Résumé. Une certaine quantité de phosphates ^3H -TdR se forme dans les cellules du méristème et les parties allongées de la racine d'*Allium cepa* durant une exposition ininterrompue de 24 h. Au cours des périodes d'observation subséquentes, le contenu de cette réserve est utilisé comme source de précurseurs pour la synthèse du DNA.

B. R. PACTER¹²

New York University,

Department of Biology, New York (N.Y., USA),

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Table II. Average grain count per square over the nuclei of the meristem region compared to that of the elongation region after exposure to ^3H -TdR for 24 h, and subsequent exposures to unlabeled medium

Amount of time in unlabeled medium (h)	Average grain count	
	Meristem	Elongation
0	3.9 ± 0.4*	6.9 ± 0.5
1	9.5 ± 1.0	11.0 ± 0.9
8	6.9 ± 0.8	8.1 ± 0.6
20	2.6 ± 0.3	3.6 ± 0.5

* Mean ± standard error.

³ M. SAMPSON and D. D. DAVIES, *Expl. Cell Res.* 43, 669 (1966).

⁴ S. R. PELC and L. F. LA COUR, *Experientia* 15, 131 (1959).

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¹² Present address: Department of Ophthalmology, New York University Medical Center, New York (N.Y., USA).

The Primexine of *Nelumbo nucifera*

The positions of apertures in the pollen wall are reportedly determined while the microspores are still enclosed within the special cell wall. In *Silene*¹, *Zea*², and *Helleborus*³, for example, the early apertures are characterized by the absence of primexine and the apposition of endoplasmic reticulum to the plasma membrane. Where the primexine fails to develop, the exine fails to form later in ontogeny. During our study of pollen development of *Nelumbo*, we found evidence which indicates a more involved method of determining apertures.

Materials and Methods. Flower buds of *Nelumbo nucifera* Gaertn. were collected from Pamplémousses Gardens, Mauritius and the anthers were dissected after 9 h and fixed in 0.2M glutaraldehyde (precipitated with barium carbonate) in 0.05M cacodylate-HCl buffer. The neutral fixative was computed to have an osmolality of 310 milliosmols⁴. The material was postfixed with 0.5% osmium tetroxide in 0.05M cacodylate-HCl buffer at

pH 7.6 brought to a calculated 230 milliosmols with glucose. The specimens were dehydrated in ethanol and embedded in Epon-Araldite⁵ using propylene oxide as an intermediate solvent. Sections were cut with a diamond knife on a Porter-Blum MT-1 microtome. Thick sections were stained with toluidine blue and mounted in anisol for phase microscopy. Unsupported thin sections mounted on 3–400 mesh grids were stained with 1% aqueous uranyl acetate and/or lead hydroxide⁶ or 1% aqueous phosphotungstic acid (pH 2.0) and examined with a Zeiss EM 9a.

Results. The primexine of *Nelumbo* consisted of fibrils or lamellae of matted fibrils aligned more or less parallel with the plasma membrane. Radially-directed posts, the probacules, penetrated from the cytoplasmic surface through the fibrillar layer to the inner surface of the special cell wall (Figure). The probacules and fibrillar matrix were always distributed over the entire microspore surface. We never saw any presumptive evidence for the