

Parthenin behaviour in different organisms

Organism tested	Phase	Parthenin concentration	Duration	Result
<i>Sclerospora graminicola</i>	Sporangia and Zoospores ^a	500 mg/1 ml 95% ethyl alcohol/ 1000 ml distilled water	1 h	After 30 min, control zoospores were released from the sporangia, swam for 20–30 min and subsequently gave rise to germ tubes, while experimental, zoospores were released from the sporangia, swam for 2–3 min but eventually disintegrated.
<i>Aspergillus flavus</i>	Conidia	500 mg/1000 mg/1 ml 95% ethyl alcohol/ 30 g maltextract/5 g peptone/15 g agar/ 1000 ml distilled water	1 week	No visual difference in growth and sporulation from the control

^aThe term sporangial germination means the release of zoospores and their subsequent germination.

repeating the experiment. The situation clearly implies that an inhibitor to an organism does not exercise the same activity in other. In *S. graminicola*, the rate of inhibition of sporangial germination by parthenin is highly significant ($p < 0.001$).

Moreover, slight antibacterial and absence of anti-leukemic activity of parthenin have already been noticed³. At the same time, response to the chemical in different organisms is seen. In this context, caffeic acid and *p*-coumaric acid, the other constituents of growth inhibitors reported in *P. hysterothorus*, have to be checked for their inhibiting potential¹.

Summary. Differential behavior of a growth inhibitor parthenin, has been observed. It inhibits sporangial germination and zoospore motility in *Sclerospora graminicola* and does not exercise the same activity in the conidial development of *Aspergillus flavus* at the same or greater concentration.

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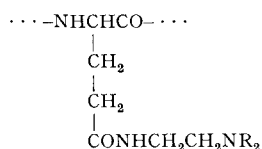
⁴ We thank Miss S. D. KANCHAN, Department of Botany, Central College, Bangalore for parthenin sample and Prof. T. R. RAMAIAH, Department of Bio-Chemistry, University of Mysore for valuable suggestions during this investigation.

Polycationic Modified Polypeptides Enhancing Poly I:C Induced Viral Resistance

It is known that viral resistance of living cells and tissues provoked by several synthetic inducers can be enhanced to a great extent by polycationic substances. DEAE-dextran was the first¹ and one of the most frequently studied and most widely used polycations. Polycationic synthetic polyamino acids like poly-L-lysine and poly-L-ornithine, as well as some other polycationic substances, have similar effects^{2,3}.

The investigation of the effect of some polycationic modified derivatives of polyglutamic acid⁴ seemed to be interesting, since some related derivatives belonging to the same group of compounds had been found to have antibacterial properties⁵ similar to poly-L-lysine, and some possibility of using them therapeutically⁶ had also been reported.

The compounds used in the present investigations were prepared from poly-methyl- α -poly-L-glutamate with 2-dialkylamino-ethylamines as described previously⁴. These polycationic macromolecules, with the following characteristic structural units as main constituents, are called poly-DMAE-glutamine if $R = CH_3$, and poly-DEAE-glutamine if $R = C_2H_5$:



The experiments were carried out in mouse L-929 fibroblast cells grown in 5% CO₂ atmosphere in Parker-199 medium supplemented with 10% of calf serum, adjusted with sodium hydrocarbonate to pH 7.2. Vesicular stomatitis virus, Indiana serotype (VSV) propagated in this culture was used for challenge. Virus assay was carried out on primary chick embryo cells by plaque titration.

The extent of the influence of polycations are expressed in 'minimal protective doses' of poly I:C ($\mu\text{g/ml}$), necessary for the complete protection of 10^6 L-cells against VSV. To determine this, 1-day-old cells were incubated at 37°C for 12 h in 1 ml serum free media containing polycation in a concentration of 20 $\mu\text{g/ml}$ and poly I:C in decreasing concentrations. After washing, the cells were challenged with 5×10^3 plaque forming units of VSV and incubated again at 37°C for an additional 48 h. The results are shown in the Table.

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The effect of the polycationic modified derivatives of polyglutamic acid was investigated on the viral resistance induced by poly I:C in primary human foetal cell cultures too; 10^6 cells were seeded in 1 ml of the above-mentioned

medium and grown in stationary tubes at 37°C for 4 to 5 days. The minimal protective doses of poly I:C, both in presence and absence of polycations, were determined as described above (Table).

Minimal protective doses of poly I:C against VSV infection in L-929 and human foetal cells in the presence and absence of polycations

Type of polycation ^b	Minimal protection doses ^a of poly I:C in $\mu\text{g/ml}$ of media	
	L-cells	Human foetal cells
Poly-DMAE-glutamine ^c	1.10^{-4}	5.10^{-3}
Poly-DEAE-glutamine ^c	1.10^{-4}	5.10^{-3}
Poly-L-lysine (Sigma)	1.10^{-3}	1.10^{-2}
DEAE-dextran (Pharmacia)	5.10^{-4}	1.10^{-3}
Without polycation	1.10^0	—

^a Determined as described in the text. ^b Polycations were used in concentrations of 20 $\mu\text{g/ml}$ of media. ^c Polycationic modified derivatives of polyglutamic acid, with characteristic structural units as indicated in the formula.

Summary. Polycationic modified derivatives of polyglutamic acid are at least as good enhancers of poly I:C induced viral resistance in various cell cultures as are DEAE-dextran or poly-L-lysine⁷.

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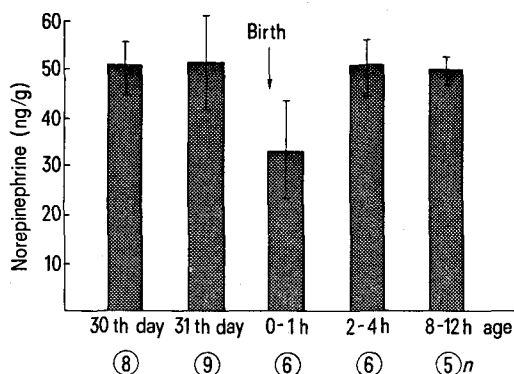
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Norepinephrine in Fetal and Neonatal Rabbit Brain

In newborn mammals, such as the rat, mouse and rabbit, the amount of norepinephrine is relatively small, slowly increasing to adult values over a period of 4 to 6 weeks^{1,2}. All these species are considered developmentally immature at birth. The newborn guinea-pig is comparatively well developed: its brain norepinephrine is at approximately the adult level. In the newborn rabbit (1-day-old) only half of this value is found³. Since we are interested in physiological modifications occurring in mammals during parturition and a few hours afterwards, we have undertaken this study to determine whether or not the brain norepinephrine store changes in fetus and newborn rabbits during this period.

Materials and methods. In our experiments we have used fetuses and newborn rabbits of the white New Zealand strain, in which the term of gestation is 31 days. At the stage of the 30th or 31st day, the female rabbits were killed by air embolism, laparotomy was performed and all the fetuses were taken out and decapitated at

once. The brains were quickly removed, blotted on filter paper and frozen immediately. Since each brain contains low amounts of norepinephrine, it was necessary to pool 4 to 5 in each sample. The newborn rabbits were separated from their mother just after birth, during the 1st h or later on, within 2 to 4 or 8 to 12 h. As with the fetuses, the newborn animals from the same litter were divided into groups of 4 to 5 and killed by decapitation. Their brains were removed and the samples were prepared in the same way. The tissue was homogenized in ice-cold 0.4 M perchloric acid by using a Tri-R tissue homogenizer (Genelab International) provided with a glass pestle, and centrifuged at $9000 \times g$ at 0°C for 30 min. The extraction was performed twice more and all 3 supernatants were pooled⁴. The pH was adjusted to 8.5 M Tris buffer⁴ and the samples were adsorbed onto alumina⁵. We used active Merck aluminium oxide, acidic activity I, prepared by the method of ANTON and SAYRE⁶. The alumina containing the adsorbed norepinephrine was washed a few times with bi-distilled water. The elution was performed with 3×2 ml of 0.3 N acetic acid, the alumina being mixed thoroughly with the acid by a magnetic stirrer. All 3 eluates were pooled, centrifuged, adjusted to pH 6.5 and used for fluorometric assay⁷. An Amico-Bowman Spectro-fluorometer with ellipsoidal mirror



Norepinephrine level in fetal and neonatal rabbit brain (ng/g). Mean values and standard error for each group are given. n = number of samples.

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