

nucleic acid metabolism should assist in elucidating the mechanism of dormancy release in light-sensitive seeds.

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### Presence of Adenosine Triphosphate in the Skin Venom of Amphibia

PRUSOFF's discovery of adenosine triphosphate (ATP) and 5-hydroxytryptamine (5-HT) in enterochromaffin cell granula<sup>1</sup> stimulated us to an attempt to extend this observation to other sections of Erspamer's enterochromaffin cell system<sup>2,3</sup>. In a preliminary investigation<sup>4,5</sup> we demonstrated the existence of ATP in "venom" squeezed manually from the parotoid glands of *Bufo vulgaris* and *viridis*, while we were unable to observe ATP in venom from *Salamandra maculosa* and *Salamandra atra* with the procedure used at that time.

This communication reports the results of investigations carried out on twenty-two species of Amphibia. The venom if possible was squeezed manually from the parotoids, otherwise ether stimulation was used<sup>6</sup>.

The presence of ATP was usually demonstrated by the method of Strehler and Totter<sup>7</sup> using diluted Sigma 'Buffered Firefly Extract'. Light emission was evaluated subjectively, or objectively by photographic and photometric means as well as by direct reaction with paper chromatograms. The photo-emission reaction was detected with a Photovolt photometer 520 coupled with a photomultiplier "1P21" and recorded with a Microcord 44 Record (Photovolt). The sample was placed in a 'Plexiglas' cylinder sealed at one end to a glass plate with chloroform and adapted to a lightproof apparatus connected by means of 'Teflon' micro tubes with two syringes which allowed the introduction of reagents. The apparatus was tightly screwed to the photomultiplier assembly. Although the instruments and the procedures used fulfil the requirements for a quantitative evaluation, we nevertheless intend to attribute only qualitative or roughly semi-quantitative values to our data because the material we were dealing with was not homogeneous. This limitation must be taken into consideration particularly when considering the "venom" collected by ether stimulation, as such "venom" contains the secretions of both granulous and mucous glands in proportions which cannot be evaluated. We also carried out investigations using paper chromatography and electrophoresis in order to demonstrate the possible linkage between oxyindolic derivatives and ATP<sup>8</sup>. As a result of our findings we were able to confirm and substantiate the luciferin-luciferase reaction.

ATP was demonstrated in the skin venom in all the species examined except for *Bombinator pachypus*. Notably, we succeeded in finding ATP in the venom from *Salamandra maculosa* and *S. atra*; techniques used on previous occasions had given negative results for these two species. From our data we therefore conclude that ATP is of usual occurrence in the "venoms" of Amphibia. Of especial interest is the observation that ATP is present in species where the "venom" does not contain 5-HT, but where other oxyindolic derivatives are present or where indolic substances have not previously been found<sup>9,10</sup>. The absence of ATP in *Bombinator pachypus*—a species with a high concentration of 5-HT in the venom<sup>9,11</sup>—may be explained because the 5-HT exists in a free form, or is bound to an unknown compound which may resemble ATP in being a nucleotide. The hypothesis that there are substances in this "venom" which inhibit the common

reaction may be discarded in view of the finding that the addition of small amounts (20γ) of ATP to the reaction mixture will rapidly produce photoemission by the "venom" of *Bombinator*.

In the species we examined variable quantities of ATP were estimated. In summary we may conclude that: (1) there is no constant relationship between ATP and 5-HT. A high concentration of ATP does not always correspond to high concentration of 5-HT and vice-versa; (2) apart from the possible relationship between ATP and 5-HT, ATP may be present in very high concentration in some species, while in others it is scarcely detectable.

The finding of ATP in yet another section of Erspamer's enterochromaffin cell system raises the possibility of being able to demonstrate the presence of ATP in other sections. Our observations are, in our opinion, particularly interesting, in view of the large variation in the composition of the granulous venom of Amphibia<sup>9</sup>. Such variability is not comparable with other sections of the system. Any interpretation of the functional significance of the ATP-5-HT linkage in the structures of the enterochromaffin cell system, however, must take into account the differences in the function of 5-HT which occur in different sections of the system.

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## GENETICS

### Age and Chromosomes

RECENTLY, I carried out chromosome analyses and sex chromatin determinations on ten patients—five females and five males—between 63 and 91 years old. The organic status of these individuals was variable, and all were ill in some way. Nevertheless, the results obtained in this investigation were reasonably uniform; their chromosomes and Barr bodies appeared normal and manifested the characteristics of the corresponding sex (Table 1).

These observations do not agree with those of Jacobs *et al.*<sup>1</sup>, who stated that the number of aneuploid cells in blood culture increases with the age of the donor, particularly above the age of 60 years. It was further claimed that these aneuploid cells lacked the sex chromosome corresponding to their sex: X in females and Y in males.

As shown in Table 2, the chromosome counts were established for 564 cells and the percentage of each was calculated. The distribution around the modal diploid number, forty-six, is similar to that found in normal cultured cells<sup>2-4</sup>. Of the 564 cells, thirty-five were karyo-

Table 1

Case No.	Sex	Age	Sex chromatin
1	♂	68	Negative
2	♀	65	Positive
3	♂	81	Negative
4	♀	79	Positive
5	♂	62	Negative
6	♀	71	Positive
7	♀	66	Positive
8	♀	91	Positive
9	♂	77	Negative
10	♂	70	Negative