

Kammerer's Alytes.

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THERE is in existence, resulting from the much-discussed experiments of Kammerer (1919, *Archiv. f. Entwicklungsmechanik*, 45, pp. 323-379, pls. x.-xi.), only a single specimen said to show the modifications described by him. This specimen is preserved in the Biologischen Versuchsanstalt, Vienna, and is the one which Kammerer recently brought with him to England and used to support his contention that he had produced heritable modifications in the midwife toad (Kammerer, 1923, *NATURE*, 112, pp. 237-238).

Although this specimen had presumably been carefully studied in England—for Kammerer (*loc. cit.* p. 237) claims: "Dozens of scientific men have seen the pads and are now convinced"—a preliminary examination of it by me in Vienna revealed such unexpected features that Dr. H. Przibram and I have found it advisable independently to make a thorough macroscopic, histological and chemical examination of the critical features of the specimen.

It will be remembered that Kammerer claimed to have produced heritable nuptial pads in a batrachian which normally lacks them. I found the specimen to have its left manus blackened both on its dorsal and ventral surfaces, the extent of the darkened area being fairly well shown in a photograph of the specimen made in Cambridge (Kammerer, 1924, "The Inheritance of Acquired Characteristics," New York, Fig. 9). A slight blackening was also to be seen on part of the right manus. Neither manus had the appearance of possessing nuptial pads, but both seemed to have been injected with a black substance, for the blackening included some of the capillaries.

An examination of the blackened areas under moderate magnification with a binocular microscope revealed that the colouring was not epidermal; that is, in epidermal spines, but in the derm. No trace of spines, points, brushes or other asperities could be seen on the integument of the prepollex, or surrounding region of either hand. The left wrist of the specimen had been lacerated. A slight pushing aside of the muscles revealed that the ventral wrist muscles and part of the palmar muscles were surrounded by a black colouring matter on all sides. This substance was in such abundance that it readily washed out in the dissecting dish water which filled the spaces between the exposed muscles. It was clear that these blackened areas were not nuptial pads; that is, patches of pigmented asperities, for the epidermis was not thrown into spines. Further, the colouring matter was not epidermal, but was distributed in great masses under the skin, between the muscles, and even in many of the capillaries on both dorsal and ventral surfaces of the left manus.

Dr. Przibram and I have independently made sections of the integument from the prepollex region of the left manus of the specimen under discussion. This is the region which in the water-breeding Salientia most frequently bears asperities. It is at the base of the first finger. Further, it was the region of maximum blackening in Kammerer's specimen. Although the specimen was poorly fixed, the epidermis in this region was intact, except along one edge where it was beginning to be shed. No suggestion of asperities are visible in my sections. The surface of the epidermis is perfectly smooth. The black colouring matter is seen to lie in great masses in the derm and among the muscle fibres, while some of the capillaries are choked with the same substance. It is therefore clear that no modifications occur in the prepollex

region of Kammerer's specimen, other than those produced by poor fixation and the black substance. Dr. Przibram has confirmed from his own sections my observations that no suggestion of asperities are present in this specimen.

The black substance, so irregularly distributed through the muscles, has the appearance of India ink, for under the highest powers the granules are black, not brownish black (or lighter) as most amphibian melanins. However, a critical test as to the nature of this substance is necessarily a chemical one. Oppenheimer (1909, "Handbuch der Biochemie des Menschen und der Tiere," Jena), in describing the properties of melanin, states that it may be changed into a lighter-coloured substance by treating it with concentrated nitric acid. Further, this product is soluble in alkalis. I have carried out a series of experiments with different kinds of amphibian integuments, some injected with India ink, and others merely fixed in alcohol or formol, and have found the test to be critical in distinguishing melanin from India ink. With Dr. Przibram's permission I removed a piece of integument from the palm of Kammerer's specimen at the base of the second finger. A large mass of black substance adhered to the dermal portions of the skin. The piece was cut into three parts and each treated for different periods, first in concentrated nitric acid, and after washing, in concentrated ammonium hydroxide. In spite of this variety of treatment, known to be critical in all cases, the black colouring matter remained intact, while the few small and widely separated melanophores readily observable under the binocular, disappeared. In this resistance to the treatment the colouring matter resembled the India ink masses in our controls. Dr. Przibram has carried these experiments further, and writes: "The black substance has also been subjected to the treatment with antiformin and withstood this reagent, which dissolves all melanins known to now." We may conclude that the substance which gives the dark appearance to the left manus of Kammerer's specimen is not melanin.

It has therefore been established beyond the shadow of a doubt that the only one of Kammerer's modified specimens of Alytes now in existence lacks all trace of nuptial pads. The question remains: Might not this specimen at one time have possessed them? There are available three kinds of evidence bearing on this point: (1) sections said to have been made from the pad of the modified specimens; (2) photographs showing more or less clearly some indication of the pads; and (3) testimony of observers who believe they saw asperities in the modified specimen.

Kammerer (1919, *op. cit.*, Pls. x. and xi.) has produced figures of histological preparations said to be made from the nuptial pad region of his specimens, or to be more exact, (translation) "through the skin of the first (inner) finger." Similar preparations were sent by Dr. Kammerer some time ago to Dr. E. Uhlenhuth, who has kindly loaned them for examination. Further, other microscopical preparations similar to those described by Kammerer are preserved in the Biologischen Versuchsanstalt in Vienna, as a material result of Dr. Kammerer's studies. Both sets of preparations agree fully with the description given by Kammerer. The controls figured by Kammerer, as well as those in Vienna and New York, exhibit a smooth epidermis, while that labelled as coming from the modified individual is thrown into numerous cornified and pigmented spines. It has been claimed that these spines are distinctive in form, and hence the pads must have been produced experimentally in Alytes.

Now, it is a well-known fact that the nuptial spines (and also the pad glands) vary enormously in size within a species. Champy (1924, "Les Caractères Sexuels," Paris, Fig. 104) has well shown how these may vary within the species *Rana temporaria*. My sections of the nuptial pads of *Bombinator igneus* differ enormously from those figured by Lataste of the same species (Kammerer, 1924, *op. cit.*, Fig. 11, sketch 8), for my animal was a terrarium animal in not very good health before it was killed. However, I have attempted in a series of forms to discover breeding pads which agreed in histological detail with the sections sent Dr. Uhlenhuth. My sections of *Bombinator maxima* agree most closely with Kammerer's sections. There is no difference in the height of the spines, their distance from each other, their form, the character of the cores within the spines, etc., between some parts of the pad in this species and parts of the sections sent Dr. Uhlenhuth. But the spines differ in proximity and height in different parts of the pads, which only goes to show that the exact form of these spines is of no value in distinguishing many species of frogs. Kammerer's sections fall within the range of variability shown by the genus *Bombinator* (more properly called *Bombina*).

The second group of evidence is to be derived from the photographs. I fail to find anything distinctive in the photograph of the specimen made in Vienna (Kammerer, 1919, *op. cit.*, Pl. x.) or that taken in Cambridge (Kammerer, 1924, *op. cit.*, Fig. 9). Dr. Przibram believes that still a third photograph, one made in Vienna (see the following note) proves conclusively that asperities were present. This photograph is not available to me, but I do not understand how asperities so small as those in Kammerer's sections would show up as Dr. Przibram points out. What proof have we that these "two or three spines" are not foreign bodies; and why do they not show up in the rather good photograph made in Cambridge (Kammerer, 1924, *op. cit.*, Fig. 9)?

In marshalling the third group of evidence, Dr. Przibram has brought together some distinguished names. With all deference to these gentlemen, I would say that the epidermis of Kammerer's specimen which is underlaid by the black substance appears, in part, slightly irregular. This appearance is probably due to the unequal distribution of the black substance below. At least, it required on my part the most careful manipulation of the lighting to prove that these irregularities were not in the epidermis. Further, I fail to see how any one qualified to pronounce on the presence or absence of nuptial pads could have examined the black discolorations on the forelimbs of Kammerer's specimen without noticing their artificial character.

A final objection which might be raised to the conclusions reached above is that some evidence has been recently presented (Kändler, 1925, *Jen. Zeitschr. f. Naturw.*, 60, pp. 175-240, Pls. 9-10) to show that the male *Alytes* may sometimes produce an incipient breeding pad. It is not my purpose to discuss whether the slight crenulations in the surface of the epidermis can be interpreted as an incipient pad. The breeding asperities Kammerer claims to have produced are true nuptial spines similar to those of *Bombinator* and cannot be compared with the epidermal irregularities of the prepolex or surrounding regions in an occasional male.

The only specimen resulting from the experiments of Kammerer has been the source of much heated argument in NATURE. By describing accurately, for the first time, the modifications in this specimen, we have proved conclusively that no pads are present. Whether or not the specimen ever possessed them is a matter for conjecture.

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It is clear from the foregoing account that the only one of Kammerer's experimentally modified *Alytes* still preserved cannot in its present state be regarded as a valid proof of the nuptial pads artificially produced in this species. We must endeavour to decide if the state the specimen is in now agrees with the state at the time of its preservation and before. The specimen is poorly fixed and preserved. Moreover, the epidermis is at several places ready to be shed or even shedding. It is a known fact, as Prof. Franz Werner, of Vienna, asserts, that during repeated handling and shaking the nuptial asperities get lost easily. The specimen has made the voyage to England and back again, and it does not look the better for it. Fortunately, there are photographic plates in existence showing the state of the specimen before it left Vienna for Cambridge and during its stay in England. One of these photographs was taken in the presence of Dr. J. H. Quastel in the atelier of Reiffenstein (Vienna), and the negative travelled with Dr. Quastel to England and has been in the possession of Mr. M. Perkins (Trinity College, Cambridge) since April 1923. A reprint of it is given in Kammerer's "Neuverbung," Stuttgart-Heilbronn, W. Seifert-Verlag, 1925 (Abb. 9, facing p. 20).

Dr. Quastel asserts in a letter to me, dated Trinity College, Cambridge, March 27, 1926, "with confidence that in the large negative of *Alytes* (taken by Reiffenstein) there are no traces of any manipulation or retouching of the actual image of the *Alytes*." Since he is not an expert in photographic matters, he has had the negative examined by an expert photographer, Mr. W. Farren, who gives the following statement: "76 Regent Street, Cambridge, March 26, 1926.—I have examined carefully the negative of *Alytes*, and while it appears to have been intensified and a string across the background retouched, there are no signs of any retouching or interference with the image of the specimen itself, or of that part of the background with which it is in contact." Mr. Perkins writes (March 20, 1926): "The only retouching which the plate has ever shown is a certain obliteration of the thread which supports the specimen, and the fact that retouching has been applied to this, in order to make it uniform with the background, enables one to state positively (from comparison) that neither the image of the specimen itself nor any point of its outline has ever been interfered with by retouching or any other process; more particularly does this apply to the external (concave) margin of the palm, where two or three spines are obviously and clearly visible. In April 1923 I had many opportunities of examining the specimen, and was always able to see the spines, whether by means of a lens or a dissecting microscope, exactly as in the photograph in question." Dr. Quastel also testifies that the copy represents the state of the specimen when it was photographed in Vienna. There seems, therefore, to be no doubt of the genuineness of the negative and photograph. Moreover, many other zoologists had examined the specimen during its stay in England, some of whom Prof. MacBride gives a list of in a recent letter to me (dated February 26, 1926, Imperial College of Science, South Kensington, London): E. Boulenger, H. M. Ververs, Cannon, F. S. Harmer, J. Stanley Gardiner, Borradaile, and F. Potts.

Further proofs that the *Alytes* in question has borne nuptial callosities in the epidermis may be found in the microscope sections of the skin that had been removed from the same specimen during the height

of the breeding season from the other (right) hand. The comparison both of Kammerer's pictures in his original paper (*A. f. Entwickl. mech.*, 45, 323, 1919, Pls. x., xi.) or in his "Inheritance of Acquired Characteristics," Boni and Liveright, New York, 1924 (Figs. 10-11, facing p. 62) and of the section photographed in America with nuptial pads of other Anura, show clearly that the callosities differ from all other known pads, resembling most those of other Discoglossidæ, as Bombinator, but still *Discoglossus pictus* (after Lataste; see Kammerer, 1924, Fig. 11 N. 3). This had already been pointed out by Mr. Perkins (*NATURE*, August 18, 1923, p. 238). Lastly, it may be mentioned that quite recently the possibility of Alytes developing nuptial pads has been shown in a specimen found in Nature and examined by R. Kändler (*Jenaische Z.*, 60, 175, 1924, Pl. 10, Fig. 12). Although it is only just a beginning of callosities in one male taken in Westfalen (Germany), when compared with the usual smooth surface of the Alytes hand it becomes significant. Kändler found the callosities in this one male on the palmar and dorsal side of the thumb, on the inner side of the third and fourth finger. The callosities resemble those which Kammerer figured for the F₂ generation of water-bred Alytes (*l.c.* Pl. xi. Fig. 4). There were no typical nuptial pad-glands. Kändler also questions if those described by Kammerer were of this character, as Kändler's females showed the same type of gland with granular content too. But no females showed callosities. Kändler has also figured the nuptial pad of other Anura, *i.e.* *Bombinator igneus* (Pl. 10, Figs. 8A, 9). This figure is in accordance with that of Lataste.

Whilst it is possible to come to a probable solution with respect to the spiculæ, we have not been able to elucidate the origin of the black substance. It is clear that it has nothing to do with the black pigment often seen in nuptial pads of Anura other than Alytes, or with the patches described and in the progeny of non-egg-carrying Alytes males by Kammerer, 1909.

The only possibility we can think of is that some one has tried to preserve the aspect of such black nuptial pads in fear of their vanishing by the destruction of the melanin through exposure to the sun in the museum case, by injecting the specimen with India ink. Kammerer himself was greatly astonished at the result of the chemical tests, and it ought to be stated that he had been asked and had given his consent to the chemical investigations. He would suggest that somebody had made such injections to get him into difficulties were it not that he remembers the black substance to have been in the same place and amount even in the living animal (letter to Przibram, February 18, 1926). So the case remains obscure.

We may conclude as a result of these observations:

(1) The only one of Kammerer's specimens of Alytes still preserved is not valid as a proof of the nuptial pads, at least not in its present state of preservation.

(2) No asperities are to be seen now, but by photographs taken before the specimen was sent to England it must be inferred that the spiculæ have been lost through the shaking of the voyage and repeated handling.

(3) Photomicrographs of the sections said to have been prepared from the skin of the same specimen some time before its preservation in the height of the breeding season show in several points different characters from nuptial pads of other Anura, even of Bombinator, coinciding more closely with those of Discoglossus (next relative to Alytes), and the picture of a rudimentary nuptial pad found on a male Alytes in Nature by Kändler (1924).

(4) The black substance in the whole specimen has nothing to do with melanin and is not restricted to a nuptial pad region.

(5) With this conflicting evidence it is greatly to be desired that the experiments of Kammerer on Alytes may be taken up again by some one of equal skill in rearing Anura.

The Progress of Geological Survey in Great Britain.

THE area described in the first¹ of the Memoirs of the Geological Survey of Great Britain referred to below lies in Ross-shire and Sutherland. The north-western corner is occupied by Lewisian, Torridonian and Cambrian rocks, all of which have been involved in the great Caledonian thrusts of the N.W. Highlands. Of special interest are the alkaline igneous rocks of Loch Ailsh. They form a composite laccolith of post-Cambrian and pre-thrusting age, made up of types ranging from perthosite (a new type composed principally of perthite) to pyroxenite, and resembling in many ways the well-known Oslo suite of rocks made classic by Brögger. Convincing evidence is adduced to show that the alkaline character is original, and quite independent of the intimate association of the intrusion with the Cambrian dolomites. Another feature of petrological importance is the description of the newer igneous rocks, which here present clear evidence of permeation and hybridisation.

The area dealt with in the second memoir² in our list, Golspie, lies to the east of the preceding, and includes the coast of Sutherland from Dornoch to Helmsdale. The earlier chapters are devoted to the physical features, and to the schists and gneisses

belonging to the Moine series and the older and newer igneous rocks associated with them. The marginal complex of injected and other metamorphic rocks around the Rogart granite is fully described, and hybrids like those alluded to above also occur. The Old Red Sandstone covers considerable areas in the sheet, and along a narrow coastal strip Mesozoic rocks are present, ranging from the trias to the Kimeridgian. The final chapter gives an account of glacial phenomena, peat deposits, and raised beaches.

Iona and Staffa are visited every summer by an increasingly large number of tourists, and those who are interested in geology will appreciate the publication of a Memoir referring to this region.³ Those who are not will become interested despite themselves, for the islands abound in geological features which are part of their attraction. The famous Ardtun Leaf Beds and Macculloch's Tree are fully described; and the columnar basalts of Fingal's Cave and other parts of Staffa are discussed on the lines followed in the recent Memoirs on the Isle of Mull.

The account of the geology of the extreme north of England given in the Memoir on Berwick and the neighbouring country⁴ is welcome as a sign that the

¹ The Geology of Strath Oyckell and Lower Loch Shin. (Explanation of Sheet 102 of the Geological Map of Scotland.) By Dr. H. H. Read, J. Phemister and G. Ross, with contributions by C. H. Dinham and M. Macgregor. Pp. vi+220+2 plates. 6s. net.

² The Geology of the Country around Golspie, Sutherlandshire. (Explanation of Sheet 103 of the Geological Map of Scotland), including a description of the Mesozoic Rocks of East Sutherland and Ross. By Dr. G. W. Lee, Dr. H. H. Read, G. Ross and J. Phemister. Pp. vi+143+1 plate. 3s. 6d. net.

³ The Geology of Staffa, Iona, and Western Mull. (A Description of Sheet 43 of the Geological Map.) By E. B. Bailey and E. M. An lerson, with contributions by G. A. Burnett, J. E. Richey, Dr. G. W. Lee, W. B. Wright and G. V. Wilson, and Petrological Notes by Dr. H. H. Thomas. Pp. iv+107+1 plate. 3s. net.

⁴ The Geology of Berwick-on-Tweed, Norham and Scremerston. (Explanation of New Series One-inch Sheets 1 and 2.) By A. Fowler. Pp. ix+58+2 plates. 1s. 6d. net.