

very slow when the satellite is far from the planet; this must indeed obviously be the case, because the tidal effects vary as the inverse sixth power of the satellite's mean distance.

- V. "On the Modifications of the Spectrum of Potassium which are Effected by the Presence of Phosphoric Acid, and on the Inorganic Bases and Salts which are found in combination with Educts of the Brain." By J. L. W. THUDICHUM, M.D., F.R.C.P.L. Communicated by JOHN SIMON, C.B., F.R.S., &c. Received March 10, 1880.

Among the results of a large investigation on which I have for many years been engaged in regard of the chemistry of the brain, I had been led to conclude that the so-called "protagon" of Oscar Liebreich is not a definite chemical body, but is a variable mixture of several bodies. This conclusion of mine (which agrees with opinions expressed on the same subject by Strecker, Diaconow, and Hoppe-Seyler) was published by me in 1874,\* and endeavours to controvert it have since then been made, on several occasions, by Dr. Arthur Gamgee.† Last summer, he brought before the Royal Society‡ his contentions for the chemical individuality of "protagon"; and it fortunately was in my power shortly afterwards to publish evidence, which, I believe, those who will take the trouble to follow it will find quite unanswerable, that Dr. Gamgee's contentions were mistaken.§ Part of my evidence to that effect consisted in showing by quantitative analyses that Dr. Gamgee's so-called "protagon" contains 0·7 per cent. of potassium; secondly, that in connexion with trifling differences in the extraction process, the proportion of potassium in different specimens of "protagon" can be made to range from a trace to 1·6 per cent.; thirdly, that with the variable quantities of potassium the quantities of phosphorus and other ingredients will also vary.

In the last published number, No. 200, p. 111, of the "Proceedings of the Royal Society," I find that Dr. Gamgee has recently brought the question again under notice of the Society, and that, in doing so, he especially rests his case upon the following statement made by his colleague, Professor Roscoe, on the subject of some examinations,

\* "Reports of the Medical Officer of the Privy Council and Local Government Board." New Series. No. III.

† "Zeitschrift für Physiol. Chemie," vol. iii, p. 260; "Ber. Deutsch. Chem. Ges.," 1879, &c.

‡ "Proc. Roy. Soc.," vol. xxix, p. 151.

§ "Annals of Chemical Medicine." Edited by J. L. W. Thudichum. Vol. i, p. 254.

which, at Dr. Gamgee's request, he had made for him: see "Proceedings," vol. xxx, p. 113:—"I have examined spectroscopically for potash a sample of protagon furnished me by Dr. Gamgee, and labelled 'Protagon, twice recrystallised, Blankenhorn.' I could not detect any potash by the spectroscope in the incinerated mass from 0.1 grm. of substance. With the carbonised mass obtained from 1.0 grm. of substance I obtained the potassium line ( $\alpha$ ) very faintly, and from comparative experiments with a dilute solution of a potassium salt I estimate the quantity of potash in 1 grm. of the substance not to exceed  $\frac{1}{20}$  mgrm. The carbonised residue of 1 grm. of protagon was carefully oxidised with pure nitric acid, when a small quantity of fused metaphosphoric acid remained after ignition. The residue weighed 0.0278 grm., corresponding to 1.08 per cent. of phosphorus.—(Signed) H. E. Roscoe."

As regards the bearing of Professor Roscoe's evidence on the question that has been raised before the Royal Society, and as regards some other relations of the same question, I beg leave to submit to the Royal Society the following observations:—

1. *On the Modifications of the Spectrum of Potassium which are effected by Phosphoric Acid.*

It is well known in spectroscopy that the best spectra of metals are obtained in the easiest manner by aid of their most volatile compounds, such as chlorides; and, on the contrary, that salts which are more or less fixed at high temperatures, such as the silicates and phosphates of alkalis and earths, give either no spectra at all or only very feeble spectra. Thus, if a minute particle of *potassic bromide* be ignited in the aerated gas flame before the slit of the spectroscope, the spectrum of potassium is at once produced in the most brilliant manner: particularly the line in red is brilliant and sharply defined, while the gas flame to the naked eye shows the violet colouring of potassium. But when a similar particle of *potassic phosphate* is ignited under similar conditions, the flame, instead of showing the violet colour of potassium, shows a greenish-yellow colour produced by volatilised phosphoric acid: and in the spectroscope, at first, only the continuous spectrum of phosphoric acid (not to be confounded with the continuous spectrum of potassium) is seen, till gradually, and at a white heat, a feeble potassium line in red, with indistinct edges, is perceived.

Even a large bead of pure potassic phosphate, when ignited before the slit of the spectroscope, never produces, even at white heat, any such intense red potassium line as the smallest bead of potassic chloride, potassic bromide, or potassic nitrate. And, in contrast with the latter salts, which volatilise quickly, the phosphate takes some time before it is completely volatilised.

When such an indolent bead of potassic phosphate which has given a feeble potassium line in red, is dipped into syrupy phosphoric acid and again ignited, the continuous spectrum of volatilising phosphoric acid is again at first obtained, the aerated gas flame to the naked eye is enveloped in a greenish-yellow mantle, and in the spectroscopic the continuous spectrum of phosphoric acid only, stretching far into the blue, and without a trace of the potassium line in red, is seen. Gradually the bead flows again as a transparent glass, and then the red potassium line reappears in the spectroscopic though without ever reaching the vivacity of the line produced by volatile salts. If now the bead have added to it a particle of a chloride—for example, zinc chloride—and be again ignited, a lively potassium spectrum will be seen in the spectroscopic, *i.e.*, the potassium goes away as chloride. That double decomposition with the separation of the products takes little more time than the volatilisation of a bead of potassic chloride of about the same size.

Evidently then, if it were intended to estimate from the spectroscopic signs the concentration of a given potassic solution, it would be necessary to take into account not only the intensity of the potassium line produced during the volatilisation of the solution, but also the length of time during which that line is perceived. If solutions equally concentrated of potassic chloride and potassic phosphate are similarly examined before the spectroscopic, the phosphatic solution is found to give the feebler band, but to give it for the longest time; and no spectroscopic observation with regard to such a solution can pretend to any validity for purposes of quantitative comparison unless the observer be equally attentive to the duration as to the intensity of the phenomenon.

Referring now, in the light of these considerations, to Professor Roscoe's letter, I wish first of all to point out that his observation, when made with the carbonised mass from 1 grm. of substance corroborated my assertion that the substance contains potassium. And I may add, by the way, that in my paper on which Dr. Gamgee comments to the Royal Society, I have given sixteen quantitative analyses of "protagon" and its congeners with regard to the quantity of potassium contained in each; having made each determination of potassium by ordinary gravimetric methods, and having weighed the potassium as platinochloride.

Secondly, however, Professor Roscoe estimates that the quantity of potash contained in a gram of Dr. Gamgee's "protagon twice recrystallised" did not exceed  $\frac{1}{250}$  mgrm., an estimate which he appears to found on the faintness of the potassium line ( $\alpha$ ) obtained by him from that substance as compared "with a dilute solution of a potassium salt." This inference I think it necessary to check by the considerations I have before stated. Professor Roscoe states that he

obtained from 1 grm. of "protagon" a residue of 0.0278 grm. of metaphosphoric acid, but according to his spectroscopic estimate, less than  $\frac{1}{20}$  mgrm. of potassium. What is the value of the latter half of this statement when taken with the context of the other half?

Before Professor Roscoe could possibly obtain from such a substance as he seems to have examined any spectroscopic evidence of the presence of potassium, he would have had to drive away from his platinum wire a relatively enormous amount of free phosphoric acid. He would then have had some potassic phosphate left, which, as we have seen, gives only a feeble and indistinct potassium band in the red, even when a large bead is heated in the flame before the spectro-scope, and which, in so extremely small an amount as he must have had, would of necessity give a fallaciously weak reaction.

Further, Professor Roscoe seems not only to have thus overlooked the inhibitive or weakening influence of phosphoric acid in relation to the spectrum of potassium, but has also omitted to give any objective measures of the intensities which he purports to compare, has moreover not recorded the length of time during which the compared phenomena continued, and has not even named what was the particular potassic salt used by him as a standard in his comparison.

Apart, therefore, from the inherently small worth of spectroscopy as a quantitative method, not even those precautions of which the spectroscopic method admitted were employed by Professor Roscoe, and I must therefore respectfully submit my opinion that his experiment is of no practical value.

## 2. *On the Inorganic Bases and Salts which are found in combination with the Educts of the Brain.*

Frémy, in his research on the brain published in 1841, stated that the body which he had isolated and termed "cerebric acid," was frequently combined with soda and phosphate of lime. Another substance, which he termed "oleophosphoric acid," he assumed to be ordinarily in the state of soda salt, but sometimes combined in part with phosphate of lime. He separated these bases and salts by dissolving the mixture of the two bodies (cerebric and oleophosphoric acid) in boiling absolute alcohol, which had been made slightly acid by sulphuric acid. Sulphates of lime and soda remained suspended mixed with some insoluble organic matter, and were removed by filtration. The cerebric and oleophosphoric acids were in solution, and deposited on cooling. From the mixture oleophosphoric acid was extracted by ether, which left the cerebric acid behind. Frémy further found that cerebric acid combined with all bases, and must be considered a true acid. When heated with dilute solutions of potash, soda, or ammonia it did not dissolve, but combined with each of these bases. The compounds were also obtained by mixing any of these

bases with an alcoholic solution of cerebrie acid. Thus, he prepared the insoluble barium compound containing 7·8 per cent. of baryta.

Bearing this experience of Frémy in mind during my researches on the chemical constitution of the brain, I invariably examined for inorganic ingredients all the educts which I obtained from the brain.\*

Of KEPHALIN (the most important of the phosphorised ingredients of the brain) 10 grms. were dissolved in one litre of water, and after filtration precipitated by hydrochloric acid. The solution of hydrochloric acid and other matters filtered from the precipitated kephalin was evaporated to dryness. A portion was then boiled with solution of baryta, when traces of *ammonia* were evolved. The rest of the residue was then ignited in a platinum dish to destroy all traces of organic matter. The ash was but slightly fusible; was only partially soluble in water, but was easily soluble in water slightly acidified by hydrochloric acid. The solution with ammonia gave an abundant precipitate of *earthy salts*, and the solution assumed a *deep blue colour*. Precipitate and solution were separated by filtration.

The precipitate dissolved readily in a little hydrochloric acid, forming a slightly red solution, suggesting the presence of *iron*, which was confirmed by the sulphocyanide test. In another portion of the solution dilute sulphuric acid gave an abundant precipitate of gypsum, showing presence of *calcium*. In another portion the molybdate test showed the presence of *phosphoric acid*. To the remaining portion a few drops of ferric chloride were added, then sodic carbonate nearly to neutrality, and, lastly, barytic carbonate. The mixture was allowed to stand, filtered, and the filtrate, free from excess of baryta by sulphuric acid, was again filtered, and the filtrate, after supersaturation with ammonia, gave a great precipitate with ammoniac oxalate, showing presence of much *calcium*. The filtrate from this calcic oxalate on concentration, and treatment with ammonia, ammoniac chloride, and sodic phosphate, gave the precipitate characteristic of *magnesium*. The precipitate produced as above described by baryta carbonate, was boiled with excess of pure soda, and the filtrate was warmed with ammoniac chloride; only a slight turbidity ensued, so that it may be assumed that *aluminium* was absent.

The *alkaline filtrate* from the foregoing precipitate by ammonia was tested for lime by oxalate, when a considerable precipitate was produced, showing the presence of *lime uncombined with phosphoric acid*, and which therefore must have been in combination with part of the kephalin. The blue solution was again filtered from calcic oxalate, which had been entirely precipitated, and was acidified with hydrochloric acid. The *copper* was precipitated by hydrothion, the filtrate

\* That part of my experience relating to kephalin and myelin is recorded in "Reports of the Medical Officer of the Privy Council and Local Government Board," new series, No. III (1874), p. 129, and *ibid.*, No. VIII (1876), p. 131.

evaporated to dryness, ignited, and the residue tested for alkalis. This residue exceeded in bulk the bases previously removed. It was fusible with ease, and on solidification became white and crystalline, but interspersed with many red particles of ferric oxide. It was a mixture of *potassic and sodic chlorides*, and of some *ferric oxide*, which, owing to the presence of the chlorides, had escaped precipitation by excess of ammonia.

It was, therefore, proved that the kephalin, obtained by the alcohol and ether process, and purified by solution in water and filtration, as described at length in my researches above quoted, is, at least in part, combined with *ammonium, sodium, potassium, calcium, iron, copper, and calcic and magnesian phosphates*. This experience was repeated a great number of times, and, in the aggregate, on several hundred grams of dry kephalin.

In the following experiments MYELIN (the phosphorised ingredient which stands next in importance to kephalin in the chemistry of the brain)\* was examined for inorganic bases, in order to obtain some knowledge regarding their relative amounts and nature. The myelin was dissolved in water, precipitated by hydrochloric acid, and the acid solution separated by filtration. The *copper* was removed by hydrothion, the filtrate evaporated to dryness, and the residue ignited in a platinum dish. During this ignition much *ammonic chloride* was given out, and its nature distinctly proved by condensing and analysing it.

The fused mass was dissolved in dilute hydrochloric acid, and the solution so obtained treated with excess of ammonia. The white gelatinous precipitate which was thrown down was scarcely coloured black by ammoniac sulphide, indicating the presence of only a trace of iron. The precipitate and filtrate were submitted to analysis, qualitative and quantitative, as follows:—

*The precipitate* was dissolved in dilute hydrochloric acid, the solution was nearly neutralised by sodic carbonate, after addition of some ferric chloride; an excess of barytic carbonate was now added; the mixture was shaken, allowed to stand, and then filtered. From the concentrated filtrate the barium was then removed by sulphuric acid, and the filtrate was precipitated in the presence of excess of ammonia by ammoniac oxalate. The oxalate so obtained was converted by intense ignition into oxide, which weighed 0.1994 grm., equal to 0.1423 grm. *calcium*. The filtrate and washings from the calcic oxalate were treated as usual for the quantation of magnesium, and there was obtained 0.5556 grm. magnesian pyrophosphate, equal to 0.1212 grm. *magnesium*. The precipitate which had been produced by ferric chloride, and sodic and barytic carbonates, was dissolved in

\* Reports, as above, No. III, p. 156.

nitric acid, and the phosphorus estimated by precipitation with ammonic molybdate, and transformation of the precipitate into magnesio-ammonic phosphate, &c. This gave 0.3266 grm. *phosphorus*, equal to 0.7480 grm. phosphoric anhydride.

The *filtrate* was proved to be free from magnesium and calcium. The potassium contained in it was extracted in the usual manner, by means of platinic chloride, giving 11.2766 grms. of the double salt ( $2\text{KCl}, \text{PtCl}_4$ ) equal to 3.651 grms. potassic chloride, or 1.911 grm. of *potassium*. The mother-liquor and washings were evaporated to dryness, and the platinum was removed by extracting the ignited mass with water acidified by hydrochloric acid. The extract so obtained was evaporated to dryness and fused; the residue weighed 3.45 grms., being pure sodic chloride equal to 1.356 grm. sodium. The quantities of sodium and potassium thus found were controlled by the analysis of a separate part of the original solution containing their chlorides; and on calculating from the residue obtained on evaporation there should have been found a total quantity of chlorides = 7.09 grms. There was found potassic chloride = 3.651, and sodic chloride = 3.450; together, 7.101 grms.

Assuming the above earthy bases to have originally existed in myelin as tribasic phosphates, and any phosphoric acid remaining over to have been in combination with potassium, then we have,

Calcium	.. = 0.1423 grm.	existing as $3\text{CaO}, \text{P}_2\text{O}_5$ .
Magnesium	.. = 0.1212	„ „ $3\text{MgO}, \text{P}_2\text{O}_5$ .
Potassium	.. = 0.5663	„ „ $3\text{K}_2\text{O}, \text{P}_2\text{O}_5$ .
Potassium	.. = 1.3447	„ } combined directly with
Sodium	.. = 1.3560	„ } myelin.
Phosphorus	.. = 0.3266	„ existing as $\text{P}_2\text{O}_5$ (0.748 grm.)

and distributed between the calcium, magnesium, and potassium.

In the kephalin research, related above, it was proved that kephalin was partly in combination with calcic oxide or lime, without any other acid being united with that base. In the myelin research it was found that, in myelin, there may be in combination even a much greater quantity of potash and soda than of lime.

A third research was now made on the mixture of phosphorised and nitrogenised substances, which Frémy termed “CEREBRIC ACID,” and which later writers have termed “PROTAGON.” This, after exhaustion with ether, was redissolved eight times in hot spirit, and after filtration reprecipitated on cooling. It was pressed to remove all mother-liquor, was then suspended in water and heated until it became thoroughly emulged with it; acid, either sulphuric or hydrochloric, was now added, whereupon the organic matter curdled and contracted; it was separated, and the acid solution was treated like the above described similar solution from kephalin and myelin. Con-

siderable quantities of the same bases and salts as those obtained from kephalin and myelin were isolated, but not examined any further.

It had been ascertained in previous operations on these mixed phosphorised and cerebrin bodies, and was now again observed, that one treatment with acid was insufficient to free them entirely from bases. The bodies which had been treated with acid were, therefore, treated with lead acetate solution in the cold, and were allowed to stand. The liquid which formed was filtered from, and pressed out of the lead-compounds, and on proper treatment yielded as follows:—*sodium*, = 1.78 grm.; *potassium*, = 1.185 grm.; *calcium*, = 0.08 grm.; and *magnesium*, 0.02 grm. The sodium was weighed as chloride; the potassium, 0.856 grm., as bitartrate, and 0.32 grm. as platinic chloride; the calcium as carbonate; and the magnesium as pyrophosphate. The respective preparations are herewith submitted for inspection.

Curious facts which deserve notice are the following. When so-called “protagon” was decomposed by baryta to obtain neurin, it was observed that the platinic chloride salt of the base always *contained potassium*. When the neurin was combined with phosphomolybdic acid, and carefully washed, and then again transformed into platinic chloride salt, it became free from potassium. In both cases the salt was crystalline, but the crystallisation was different in the two cases. The crystals containing potassium were most perfectly formed individuals, sometimes more than half an inch in length and breadth, and an eighth of an inch in thickness, while, on the other hand, the crystals free from potassium were bundles of needles, of lighter aspect and not admitting of angular measurement. The experience shows that the platinic chloride compound of potassic chloride is dimorphous, and in one of its forms is isomorphous with the platinic chloride salt of neurin hydrochlorate. It is further remarkable that the impure isomorphous mixture crystallised in well-defined individuals, while the pure salt crystallised in confused masses and in bundles of needles. The crystals of the perfect type, when powdered and burnt on a platinum wire in an aerated gas flame before the spectroscope, produced the line in red characteristic of potassium. This specimen is herewith submitted for inspection, as also a specimen of the salt free from potassium.

The foregoing data may perhaps contribute to explain some of the discrepancies which have been found between different specimens of neurin platinic chloride, and have caused the inconvenience of a number of different formulæ being attributed to neurin, namely,  $C_5H_{15}NO_2$ , or  $C_5H_{14}NO$ , or  $C_5H_{14}N$ , or  $C_5H_{12}N$ . They may also explain why neurin from “protagon” has been supposed to consist of a mixture of two bodies, having respectively the formulæ  $C_5H_{15}NO_2$ , termed oxyneurin, and  $C_5H_{15}NO$ , termed neurin. According to my own numerous analyses (Reports as above, No. VIII, p. 118, *et seq.*), the



principal ammonium base from the phosphorised substances has, after purification by the phosphomolybdic process and combination with hydrochloric acid and platinic chloride, invariably the composition  $C_5H_{13}NO$ .

The stoichiometric relations of such a base, in its platinic chloride salt, would be doubly disturbed by the presence of potassium; for not only would a portion of the platinic chloride, namely that combined with potassic chloride, be erroneously referred to neurin, but the entire amount of platinum, if estimated, as is usual by combustion, would be raised in apparent amount by the presence of the potassic chloride as an invisible impurity.

The ALBUMINOUS INGREDIENT of the brain contains considerable amounts of the usual salts, particularly calcic phosphate, of which the calcium is obtained as gypsum, when the albumin is treated with dilute sulphuric acid, and the solution of hemi-albumin is filtered from the undissolved hemi-protein and slowly evaporated. By combustion of the albuminous matter of the brain, all the usual salts and bases, including copper, iron, and manganese, are liberated.

The greater part of the *soluble salts* contained in the brain passes into the water extracts, while the greater part of the *insoluble salts*, such as phosphates of earths, remain with the albumin; but a portion of both remains with the phosphorised and nitrogenised principles, and follows them into all their solutions in ether and alcohol. Only *sulphates* have not been found in these latter, though they are present in the water extracts.

It follows that all educts from the brain have to be freed from the salts or bases described before they can be subjected to quantitative chemical examination; and an error which seems to me to pervade all analyses of the brain for inorganic ingredients hitherto is the following. When the brain is burnt in its entirety, the phosphoric acid resulting from the destruction of the phosphorised compounds expels sulphuric, hydrochloric, and carbonic acids, and comes to be considered as an inorganic ingredient, the fact of its previous combination being overlooked. When the brain is freed from matter soluble in spirit and ether, and the albumin and water extract alone are examined for inorganic ingredients, the fallacy otherwise introduced by the phosphoric acid in organic combination is no doubt avoided; but the inorganic salts described above, as in combination with the phosphorised and nitrogenised matters are overlooked and left out of consideration.

A vast range of other organic analyses (animal and vegetable) for inorganic constituents is no doubt affected by the same fallacy, and it seems to me that the right making of such analyses is a much more complicated problem than has hitherto been believed.