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ON THE FORMATION OF FAT IN THE ANIMAL BODY[1]. By J. B. LAWES, M.D., and J. H. GILBERT, M.D.

PLATE XXII.

FORMERLY it was supposed that the fat of the Herbivora was derived exclusively from ready-formed fat in their vegetable food. Liebig showed that this could not be the case; and he attributed much of the fat of the animal body to the carbohydrates of the food. His views on the point were at first called in question by Dumas, Boussingault, and others, but afterwards accepted. Our own very numerous feeding experiments, commencing about 30 years ago, together with a careful consideration of the experience of practical feeding, afforded strong confirmation of Liebig's conclusions; and more especially in 1852[2], and subsequently, we pointed out the bearing of the results on the question.

At a meeting of the Congress of Agricultural Chemists held in Munich, in 1865, Professor Voit combatted this view. From the results of experiments with dogs, made in Pettenkofer's respiration-apparatus, he maintained that fat must have been produced from the transformation of nitrogenous substance; and further, that this was probably the chief, if not the only, source of the fat, even of Herbivora. In 1869 he elaborately argued the point, not only in reference to the results of his own experiments with dogs and with cows, but also to the records of those of various other experimenters, with various descriptions of animal; and he has subsequently made further contributions on the subject, conjointly with Professor Pettenkofer. If their results, obtained with a dog, and their conclusions

[1] The substance of this paper was given by one of us at the Naturforscher Versammlung (Section für Landwirthschaft- und Agricultur-Chemie) at Hamburg, in September, 1876.

[2] On the Composition of Foods in relation to Respiration and the Feeding of Animals. *Report of the British Association for the Advancement of Science, for* 1852.

drawn from them, were to be described in a few words, they might perhaps be so as follows:—When a dog was fed on starch or sugar, alone, or with albumin, or with fat and albumin, the carbon stored up was not more than that in the fat of the food *plus* that which could be derived from the albumin broken up. There was, therefore, no proof that fat can be formed from the carbo-hydrates. Again, when a dog was fed with starch and a little fat, but no albumin, the carbon stored up was equal to that of the fat of the food *plus* that derived from the transformed nitrogenous substance of the body. More starch reduced the amount of carbon stored up; the carbo-hydrate having protected the albumin of the body from oxidation, and thus limited the formation of fat. They never found fat formed from starch or sugar. They maintain that the same must occur with the Herbivora; and that to establish the formation of fat from the carbo-hydrates, experiments must be brought forward in which the fat deposited is in excess of that supplied by the food *plus* that which could be derived from the transformed albumin.

This, many of our own experiments with pigs do clearly show; and in 1866 we published a short paper[1], in which we illustrated the bearing of some of them on the point. In his paper in 1869[2], Professor Voit quotes some of those results, and admits that in the experiments in which there was only a medium albuminous supply in the food, there was, as the figures stand, a considerable deficiency for the formation of the fat produced, and a greater deficiency still in the cases in which the relation of the nitrogenous to the non-nitrogenous constituents was such as experience has shown to be the most favourable for pig-fattening; and that, therefore, a considerable amount of fat would, in these instances, appear to have been derived from the carbo-hydrates. Still, Professor Voit says he cannot allow himself to consider a transformation of carbo-hydrates into fat to be proved thereby. He confesses that he has not been able to get a general view of the experiments out of the mass of figures recorded, and suggests several possible sources of error, his reference to some of which shows that he has in fact misunderstood them. At the same time, he proposed that new

[1] On the Sources of the Fat of the Animal Body. *Philosophical Magazine*, December, 1866.

[2] *Zeitschrift für Biologie*. Band v.

experiments with geese and with pigs should be made, in order to arrive at a final decision on the question; and in a very recent conversation with one of us, he expressed his willingness to undertake a conclusive experiment with pigs.

Weiske and Wildt[1] undertook an investigation to determine the point; which, from a theoretical point of view, was well conceived; but which did not succeed, owing to the oversight of the conditions indicated by experience as essential to the rapid fattening of the animal. They selected four pigs; two were slaughtered to determine the initial composition; one was fed on food so rich in nitrogen that it suffered in health, and the experiment had to be discontinued; the other was fed on food so poor that it fattened extremely slowly; and hence, at the conclusion, calculation showed that there was enough of the consumed nitrogenous matter available for fat-formation to cover the whole of the fat which had been produced.

Thus, it has been concluded from experiments with animals which are not preeminently fat-producers, that fat is probably never formed in the body from the carbo-hydrates; and some of the experiments with more suitable animals have been, to say the least, inconclusive. Further, it seems to be assumed, that no absolute proof on the point can be obtained without the aid of a respiration-apparatus. These views, moreover, have already been adopted, not only by some physiologists, but in some text-books treating of the application of chemistry to the feeding of the animals of the farm. Thus, Professor Emil Wolff, in his *Landwirthschaftliche Fütterungslehre*—although he admits that the amounts of increase produced in relation to constituents of food consumed, which it is established by common observation may be obtained with pigs, and still more those recorded in some direct experiments with those animals, are almost incomprehensible without assuming the direct concurrence of the carbo-hydrates in the formation of fat—nevertheless, seems to consider that evidence of the kind in question, and we suppose our own, therefore, is inconclusive. He says that exact experiments are still wanting; and he suggests that accurate respiration-experiments with pigs should be made, to settle definitively whether the carbo-hydrates, as well as albumin, can contribute directly to the formation of fat in the animal body.

[1] *Ib.* Band x.

Since the appearance of Professor Emil Wolff's work, and the publication of the negative results of Weiske and Wildt, we have carefully reviewed and recalculated many of the results of our feeding experiments, including those with oxen, with sheep, and with pigs; in order to satisfy ourselves whether any doubt could be entertained of the views we have previously advocated; and whether, therefore, it was at all incumbent upon us to institute new experiments on the point. The result of this examination, so far as the ruminant animals are concerned, has been to show that, owing to the comparatively small amount of increase obtained with them from a given amount of constituents consumed, the quantity of nitrogenous substance passed through the system for the production of a given amount of increase was, in most, if not in all cases, so large as, in the absence of proof to the contrary, to admit of the assumption that the whole of the fat formed had its source in transformed nitrogenous matter. At any rate, no absolute proof of the derivation of fat from the carbo-hydrates can be obtained from data of the kind in question relating to such animals. In deciding the point in regard to them, the evidence afforded by the analysis of the fæces and of the urine, and by the determination of the products of respiration, must also be brought into consideration. It was quite otherwise, however, in the case of our experiments with pigs; in many of which much more fat was produced than could possibly have been derived from transformed albumin of the food. We concluded, therefore, that we were in no way called upon to institute new experiments, and decided, instead, again to direct attention to the results quoted in the short paper on the subject published in 1866, as already referred to.

The figures given in Table I. of that paper show how much smaller is the proportion of alimentary organs and contents in a given live-weight of the pig than of either oxen or sheep; that, in proportion to a given live-weight, the pig consumes a very much larger quantity of dry substance of food within a given time (whilst his food contains a very much larger proportion of digestible, and therefore, very much less of necessary effete matter); that he gives several times as much increase in relation to a given live-weight within a given time; much more increase in relation to a given quantity of dry substance

of food; also a larger proportion of fat in that increase. Further, the most appropriate fattening food of the pig contains a larger proportion of readily digestible carbo-hydrates than that of the ruminant animals. All these conditions indicate the pig to be the most suitable animal for the determination of the point in question.

The results selected to illustrate the main point are given in Table II. of the same paper. They were all obtained more than 20, and some more than 25 years ago; and the rations were not arranged with a special view to the settlement of this question; but to determine the relations of the different constituents of food to various exigencies of the body, and the amount, and the proportion of different foods which were the most favourable for the feeding of the animals. Accordingly, the series included proportions varying from 2·0 to 6·6 parts of non-nitrogenous to 1 of nitrogenous substance in the food.

In experiment 1, two animals were selected, of the same litter, and as nearly as possible alike both in character and weight; the weight of the one being 100 lbs., and that of the other 103 lbs. One was slaughtered at once, and its contents of nitrogenous substance, fat, mineral matter, &c., accurately determined. The other was fed on a mixture consisting of bean-meal, lentil-meal, and bran, each one part, and barley-meal three parts, given *ad libitum*, but accurately weighed, for a period of ten weeks, when it had nearly doubled its weight. The food contained, however, a considerably higher proportion of nitrogenous to non-nitrogenous constituents than is recognised as the most favourable for the fattening of the pig. The animal was then slaughtered, and analysed, as the other had been. The composition of the food was also determined by analysis. The experiment afforded, therefore, reliable data for determining the amounts of fatty and nitrogenous substance consumed, the amount of nitrogenous substance stored up in the animal as such, and also the amount of fat stored up.

Eight other experiments were quoted, in each of which a different food-mixture was employed, and in each of which three animals were fed, in some cases for a period of eight, and in others of ten weeks. The average live-weight per head at the commencement was, in these eight experiments, respec-

tively, 143, 147, 144, 149, 95, 95, 94, and 97 lbs. Thus, in the first four cases, the average initial weight per head was notably more than that of the two animals of experiment 1; but in the last four experiments it was very nearly the same. In the calculations, the percentage composition of the animals in experiments 2—9 was assumed to be the same at the commencement as that of the unfattened animal in experiment 1, and the same at the conclusion as that of the fattened animal in experiment 1. It was quite obvious, during the progress of the experiments, that the animals having the higher proportions of nitrogen in their food, grew more, and fattened less, than the others; and careful observations, made after slaughtering, entirely confirmed this. The tendency to error in the calculations would be to indicate too low an amount of nitrogenous substance, and too high an amount of fat stored up in the cases with the higher proportions of nitrogenous substance in the food, and too high an amount of nitrogenous substance, and too low an amount of fat stored up with the lower proportions of nitrogenous substance consumed. The range of the probable error here supposed is, however, not such as at all to throw doubt on the validity of the main conclusions which are drawn from the figures as they stand.

A comparison of the amount of ready-formed fat in the food, with that of the determined or estimated total fat stored up in the increase of the respective lots of animals, showed that, even supposing the whole of that consumed had been retained, there remained from two-thirds to nine-tenths of the total amount stored up to be otherwise accounted for. It must have been produced within the body.

The next question was, whether this large amount of produced fat could possibly have been derived from the nitrogenous constituents of the food? or whether it must of necessity have had its source, in greater or less proportion, in the carbohydrates at the same time supplied?

Deducting from the total amount of nitrogenous substance consumed, the small amount estimated to be stored up as such in the increase of the animal, there remained a large proportion available, it may be, for the formation of fat, with other products. In order to give to the nitrogenous substance of the

food not stored up, its fullest possible (and even more than its fullest) value for fat-formation, the whole of its carbon, *minus* that which its nitrogen would require to form urea, is, for the sake of illustration, assumed to be available for fat-formation.

So calculated, the result in experiment 1, and also in two of the other cases in which the proportion of nitrogenous to non-nitrogenous substance in the food was considerably higher than is recognised by experience as the most suitable in the fattening food of the pig, was that more nitrogenous substance was available for fat-formation than was necessary to supply the estimated amount of produced fat. In the cases in which the nitrogenous substance was not so excessive, but still more than is the most appropriate, there was a considerable proportion of the total produced fat which could not possibly have been derived from the nitrogenous substance of the food. Lastly, when the proportion of the nitrogenous to the non-nitrogenous substance in the food was the most appropriate for fattening, there was a much larger proportion (about 40 per cent.) of the total produced fat, which could not possibly have had its source in the nitrogenous substance consumed.

Striking as are these results, it is obvious that a still larger proportion of the produced fat would appear to be formed from the carbo-hydrates, if it were assumed, with Henneberg and Voit, and as is doubtless nearer the truth, that 100 parts of albumin will not yield more than 51·4 parts of fat, instead of, according to the foregoing illustration, about 61 parts.

It will be well, however, briefly to consider, whether an amount of error in the estimates, which would turn the scale, and show that the whole of the produced fat might be derived from the nitrogenous substance of the food, is at all conceivable, at any rate in the cases in which the proportion of the nitrogenous to the non-nitrogenous constituents consumed was the most nearly that which is recognised as the most favourable for pig-fattening, and in which the largest amount of formation from the carbo-hydrates is indicated.

Obviously, the most important point to consider is the range of error admissible in the estimation of the fat stored up in the increase of the animal.

It would be necessary to reduce the estimate of the amount of fat stored up by more than 30 per cent. to bring it low enough to be covered by the fat in the food, *plus* that derivable from the transformed nitrogenous substance, leaving all the other calculations the same. If, however, we were to assume that 100 nitrogenous substance yielded only 51·4 fat, it would be requisite to reduce the estimate of the fat in the increase by more than 40 per cent., to reverse the indication. This is on the assumption that the whole of the fat of the food was stored up in the animal, which would certainly not be the case. It is also on the assumption that the whole of the nitrogenous substance of the food, not stored up as such in the increase, was digested, and available for transformation into fat, &c., but this again is certainly not the case. According to our own experiments, it may be supposed that, with a pig feeding exclusively on good barley-meal, about one-sixth of the total nitrogen voided would be in the fæces. But if it be assumed, according to the estimates of E. Wolff[1], that 20 per cent. of the nitrogenous substance, and 32 per cent. of the fat of the barley, would be voided undigested, and therefore without contributing to the deposition of fat, our estimate of the amount of fat stored up in the increase would have to be reduced by more than 55 per cent., or considerably more than half, to bring it within the amount derivable from the resorbed fat, and the transformed nitrogenous substance of the food.

It is submitted that a range of error in our estimates, at all approaching even the lowest of those above assumed for the sake of illustration, is simply impossible. It is further submitted, with the utmost confidence, that such is the wide margin in the case of pigs fattening rapidly on their most appropriate fattening food, that the question of whether or not the carbo-hydrates contribute to fat-formation may be conclusively settled by a properly conducted experiment with those animals, without any analysis of the fæces or the urine, or any determination of the products of respiration. To this end, we would suggest that two animals be selected, of a breed of good fattening quality, and as nearly as possible alike in characters and in weight. A convenient size and weight would

[1] *Landwirthschaftliche Fütterungslehre*, Appendix, Table I.

be, say about 90 lbs. per head. Let each be fed with ground barley of good quality, giving it, by degrees, as much as it will consume, until both weigh about 100 lbs. Then slaughter one, and determine its total amount of nitrogenous substance, fat, &c. Feed the other in the same way, that is with barley-meal (and water) exclusively, as much as it will consume, until it reaches about 200 lbs. in weight. Then slaughter and analyse it, as the first. The quantity and composition of the food must, of course, also be determined. Such an animal would consume somewhere about 500 lbs. of barley, more or less, and increase from 100 lbs. to 200 lbs. in live-weight, in from 8 to 10 weeks, more or less, according to quality of the animal, quality of the food, &c. &c. It is desirable that the animals selected should have been feeding on fairly good food previously, so that the transition to full fattening food should not be too sudden. It is also, of course, desirable, that the experiment should be made in duplicate if possible.

But, independently of the results of any such experiments, it may be asked, what is the lesson of common experience in this matter? We say, unhesitatingly, that the experience of the feeding of animals fully confirms our view.

In reference to this point we would call attention to the coloured diagrams Pl. XXII. which show the proportions of nitrogenous substance (black), of non-nitrogenous substance (yellow), and of total organic substance, nitrogenous and non-nitrogenous together (blue), respectively:—

I—consumed per 100 lbs. live-weight per week,

II—consumed to produce 100 lbs. increase in live-weight,

in the case of thirty different feeding experiments with pigs, each of which comprised not less than three and some four animals, and in each of which they fixed their own consumption[1]. That is to say, various current foods, but containing widely different percentages of nitrogenous substance, being selected, one (or a mixture) of high, or of medium, or of low

[1] "Pig Feeding;" *Jour. Roy. Ag. Soc. Eng.* Vol. XIV. Part 2; Experiments 1—8, and 12, Series 1; Experiments 1—12, Series 2; Experiments 1—5, Series 3; also "On the Equivalency of Starch and Sugar in Food," *Report of Brit. Ass.* for 1854; Experiments 1—4. See also "Experimental Enquiry into the composition of some of the Animals fed and slaughtered as human food." *Phil. Trans.* 1859, Part 2.

percentage of nitrogen, was given, *ad libitum;* or a fixed quantity of one or more was given, and another given *ad libitum;* and so on. In this way the animals fixed their own consumption, and from the results it may be judged by what requirement this was guided.

First, as to the consumption by a given live-weight within a given time; which of course met the collective requirements for both sustenance and increase. Diagram I illustrates this point. The lowest amount of nitrogenous substance so consumed in any one of the thirty experiments is taken as 100; and it is seen that the amount of it consumed ranged, among the thirty dietaries, from 100 parts to more than 300; and it averaged more than 200. Reckoned in the same way, the consumption of non-nitrogenous substance varied from 100 to only 177 parts, and averaged only 141 parts. Again, reckoned in the same way, the consumption of total organic substance (nitrogenous and non-nitrogenous together) ranged from 100 to only 150 parts, with an average of 125 parts.

Secondly, as to the amounts consumed to produce 100 lbs. increase in live-weight. Diagram II shows that, for this result, the consumption of nitrogenous substance ranged from 100 to 282 parts; and it averaged 173 parts. That of the non-nitrogenous substance ranged from 100 to only about 140 parts, with an average of 124 parts; and that of the total organic substance (nitrogenous and non-nitrogenous together) from 100 to only 147 parts, with an average of 122 parts.

It should be explained that, as in the Tables and Diagrams given in the original papers above referred to, the *total* amounts of nitrogenous and of non-nitrogenous substance, in the different foods, are taken as the basis of the calculations; no deduction being made for "indigestible" matter; nor is the fat in the food reckoned at any higher value than the other non-nitrogenous constituents. This plan was adopted as best representing the facts actually determined by analysis; but attention was at the same time directed to the varying amounts of indigestible matter in the different foods, and to the greater or less amounts of fat which they contained. We have, however, quite recently recalculated the whole of the experiments, making deduction for indigestible or undigested matter, according to E. Wolff's

Table already quoted, and with him multiplying the amounts of fat by 2·5, and have constructed Diagrams according to the data so obtained. These still more strikingly illustrate the point in question than the Diagrams herewith given ; that is to say, they show a wider range in the amounts of the nitrogenous substance consumed in the different experiments, a less variation (excepting in one case in which there was much fat) in the amounts of the non-nitrogenous substance consumed, and especially a less range in the amounts of the total organic substance consumed. The two methods of calculation show, however, in most of the cases, much less difference in the relation of the nitrogenous to the non-nitrogenous constituents than might have been anticipated. With this explanation, we still adhere to our original plan of calculation, rather than adopt corrections based upon factors as yet not sufficiently established[1]. At the same time, we repeat that the points here indicated should be considered in judging of the results as they stand.

It is then perfectly clear, that neither the amount of food consumed in relation to a given live-weight within a given time (which of course covered the requirements for increase as well as sustenance), nor the amount taken to yield a given amount of increase in live-weight (which in its turn covered the requirements for sustenance also), was at all in proportion to the amount of the nitrogenous constituents it supplied. It is quite obvious, that the consumption, both for sustenance and for increase, was much more nearly in proportion to the amount of digestible non-nitrogenous constituents supplied; but it was more nearly still guided by the amount of the total digestible organic substance—nitrogenous and non-nitrogenous together—which the foods contained.

That the great variation in the amount of nitrogenous substance consumed was not due to a deficiency of it in most of the foods employed, is shown by the fact that it was in the experiment in which the food contained the lowest proportion of it, that the smallest amount of nitrogenous matter was not only consumed in relation to a given live-weight within

[1] Professor Emil Wolff has recently determined the proportions undigested of the different constituents of cocoa-nut cake, barley-meal, maize-meal, and pea-meal, in actual experiments with pigs. *Versuchs-Stationen Organ.* Band XIX. No. 4, 1876.

a given time, but was required to produce a given amount of increase. It is obvious, that where two or three times as much nitrogenous substance was consumed, it was much in excess of the normal requirement. In fact, the animals consumed almost regardless of the amount of nitrogenous substance supplied, until they had obtained a sufficiency of non-nitrogenous, or of total organic substance. It is further obvious, that the range of variation in the amounts of non-nitrogenous constituents consumed would have been very much less, but for the very variable amount of nitrogenous substance necessarily taken with it, the variable amounts of fat in the foods, and the greater amount of indigestible matter in some of them than in others. The indication is, indeed, that the excess of nitrogenous substance consumed substituted a certain amount of non-nitrogenous constituents; that, in fact, within certain limits, the two classes of constituents may, for the purposes of respiration and fat-formation, mutually replace each other.

Lastly on this point, not only did neither the amount of food consumed, nor the amount of increase in live-weight yielded, bear any relation to the amount of nitrogenous substance supplied, but the more excessive the supply of it the greater was the tendency to grow, and the less the tendency to fatten. There is, of course, a point below which the proportion of nitrogenous substance in the food should not be reduced; but if this be much exceeded, the proportion of the increase, and especially of the fat-increase, to the nitrogenous substance consumed, rapidly decreases; and it may be stated generally, that taking our current fattening food-stuffs as they are, it is their supply of digestible non-nitrogenous, rather than of nitrogenous constituents, which guides the amount, both of the food consumed, and of the increase produced, by the fattening animal.

In conclusion, we repeat that, in many of our experiments with pigs, much more fat was produced than could possibly have been derived from the albumin of the food, and hence the carbo-hydrates must have contributed directly to its formation; further, that experience in practical feeding is entirely in accordance with our views on the point.

DIAGRAM I SHOWING THE PROPORTIONS CONSUMED PER 100 LBS. LIVE-WEIGHT PER WEEK.

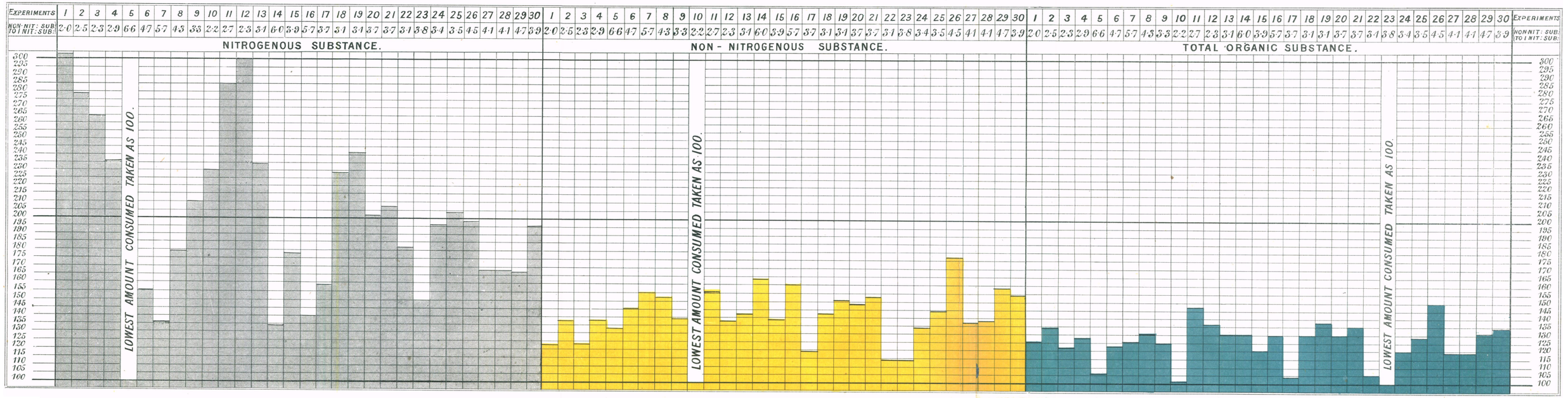

DIAGRAM II._ SHOWING THE PROPORTIONS CONSUMED TO PRODUCE 100 LBS. INCREASE IN LIVE WEIGHT.