

THE PROBLEM OF NITROPHILY*

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INTRODUCTION.

A plant association according to BRAUN-BLANQUET and PAVILLARD is a "community characterised by its essential homogeneous floristic composition, at least as regards the dominant species and recognised and characterized by its specific assemblage and principally by its *characteristic* species." This definition was accepted by the Sixth International Botanical Congress in 1935.

The concept of the *characteristic* species is very important, and has considerably added to the accuracy of the definition. Probably it has been influenced by the work of UNGER (1836) since plants belonging to the third group in his classification namely "Restricted to soil" can be the characteristic species of BRAUN-BLANQUET. The fundamental idea behind this concept is that there are certain species of a plant association that are so fastidious regarding their habitat conditions that they cannot grow in any other habitat but that particular one. Thus in the case of equal commensals at least, the characteristic species must be those species which can only grow under a particular edaphic or micro-climatic factor which predominantly governs that association. The consistent presence of such plants in certain well defined habitats along with their conspicuous absence from all other associations, has led workers to term them "Indicators". A reliable indicator of certain conditions of habitat can from its presence and other sociological data enable the worker to understand and appreciate the habitat quickly and effectively. Plant indicators have thus been used extensively in Europe and America to the betterment of their agriculture and silviculture. This more or less rigid limitation of the plant to definite plant communities depends upon its degree of fidelity which is measured statistically as a synthetic character.

We are of the opinion, however, that mere statistical data, though formidable are not sufficient, since floristic composition as admitted by BRAUN-BLANQUET himself (1932) is vague. According to him no two bits of vegetation have precisely identical floristic combination. Thus in addition to statistical data an attempt should be made to get a more direct proof from the habitat itself.

The control of the habitat is complex and difficult in the case of unequal commensals but in the case of equal commensals where all the plants depend upon a single factor in the soil, like the nitrate factor for instance, the task becomes much easier. This factor has been studied by various workers like BAUER, CARSTEN OLSEN, WEBER and many others.

Though generally speaking the nitrates are essential to plants for the synthesis of proteins and their absence from the substratum causes malnutrition, very few plants as such can grow favourably in a high nitrate habitat. The nitrate content of the soil varies from habitat to habitat. In moist waterlogged places for instance, the nitrate concentration is very low, whereas in dung heaps, manure beds and such other places where organic remains of plants and animals are allowed to decay, it is considerable.

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It is found from observations that such habitats have a particular type of vegetation, which differs from other plant associations growing on moderate or low-nitrate habitats. This particular type of vegetation, so characteristic of the habitat, can therefore be termed nitrophilous.

We have, therefore, set out along a new line of approach, viz. a study of the Nitrophilous Vegetation according to both chemical and statistical data. The results have been synthesised according to a new formula proposed by us and from its use we have produced a list of a few Indian nitrophilous plants and have graded them according to their nitrophily and indicator value. Some of them are so specific about their habitat conditions that they may be termed the characteristic species of the nitrophilous association.

DEFINITION.

Probably the oldest definition of nitrophily was that advanced by SCHIMPER (See JACKSON 1928) who defined nitrophilous plants as alkali lovers. It becomes difficult to understand this definition. Possibly, it may be because of the belief that nitrification cannot take place in acid soils. This hypothesis has since been disproved by the work of CUTLER (1931), who discovered an organism which can nitrify in an acid medium and thus enunciated that nitrification can and does take place even in a medium with pH 4.5. It is, therefore, regrettable that this outdated definition has appeared in the "Ecological Glossary" compiled by J. RICHARD CARPENTER as late as 1938, in spite of the fact that better and more precise ones are now available.

The term 'Nitrophily' literally means love for nitrogen. A nitrophilous plant is one to which a high-nitrate concentration is necessary. Hence it thrives better in a high-nitrate habitat than in any other and nearly always occurs in such habitats in contrast to other plants which cannot tolerate such a high concentration of nitrates.

On the other hand a more passive view is also possible viz. that nitrophilous plants possess certain properties within them which enable them to merely tolerate a high nitrate habitat better than many other plants.

This view though also in keeping with the definition of love for nitrogen is in a way very much different from it and completely changes the whole aspect of the situation. Thus we have in a sense two views diametrically opposite: nitrate factor as a rigid necessity for nitrophilous plants on the hand, and on the other hand a mere capacity to tolerate nitrates. The question then arises, which of the two views is the correct one and upon its answer hangs the whole problem of nitrophily.

We shall try to answer the question by referring to various workers and the definitions which some of them have advanced.

WARMING (1925) has defined nitrophilous plants as those plants which thrive best in soils where compounds of Ammonium and Nitric Acid are abundant and therefore in the vicinity of human dwellings. He further stated that they belong to special families like *Chenopodiaceae*, *Cruciferae*, *Solanaceae* etc. and that other species develop feebly on such soils because they take into their tissues more nitrates than they can endure.

BAUER (1938) in his work on the city dumps of Leipzig-Moekern has found that nitrates are stored in mature plants in decreasing amounts in the following species: *Amarantus retroflexus*, *Chenopodium album* and *Atriplex nitens*. He further stated that in sand cultures the following plants could endure high concentrations of nitrates: *Amarantus retroflexus*, *Atriplex nitens*, *Hyoscyamus niger* and to a lesser degree, *Fagopyrum esculentum* and that *A. retroflexus*, *Ballota nigra*, *Hyoscyamus niger* and *Solanum dulcamara* are not obligate dump dwellers.

CARSTEN OLSEN (1921) in his "Ecology of *Urtica dioica*" states that *Urtica dioica*

belongs to the so-called "Nitrate-Plants" which greedily absorb nitrates from the soil and whenever not directly assimilated, they are accumulated as reserve material in plant tissues which could be shown by micro-chemical tests with Diphenylamine-Sulphuric acid. He further states that plants showing nitrates in their tissues can only have absorbed them from the soil since plants cannot transform other nitrogenous compounds to nitrates and that plants frequently growing along with *U. dioica* like *Mercurialis perennis*, *Stachys sylvaticus* and *Chrysoplegium alternifolium* also show a high-nitrate content. Thus a rich growth of *U. dioica* always coincides with an intensive capacity of nitrification in the soil.

According to BRAUN-BLANQUET (1932), intensive nitrification occurs in most meadow soils and in many forest soils with a herb layer. Individual communities such as *Alnus* woods with numerous herbs, and deciduous forests in Sweden, also communities around springs are characterized by a high nitrate-content. HESSELMAN (1917) found snow-clad communities composed of *Catabrosa algida*, *Poa alpina*, *Cerastium cerastioides*, *Saxifraga stellaris* and other species rich in nitrates.

BRAUN-BLANQUET also mentions that halophilous shore vegetation composed of *Atriplex* and *Honckenyia peploides* also give a strong nitrate reaction and further states that nitrophilous plant communities are distributed in dry subtropical regions, the most nitrophilous community of the Mediterranean region being the *Silybum-Urtica pilulifera* association of which all species show a high nitrogen-content when young.

From the study of nitrophilous lichen associations of bird roosts and marmot rests, of stones and rocks wet with goat and sheep urine, SERNANDER (1912), FREY (1922), GAMS (1924) and MOYKA (1924) (see BRAUN-BLANQUET 1932) have found that nitrophilous rock-lichen communities are often arranged in a distinct belt-like order according to their degree of nitrophily. Upon the bird roosts and marmot rests of the Alps and the Tatra, the surfaces of overmanured rocks are occupied by the extremely nitrophilous *Ramalinetum strepsilis*, with *Rinodina demissa*, *Xanthoria fallax* and *Physcia tribacia* as characteristic species. On sloping surfaces where nitrogen compounds are washed by rain-water grows *Alectorietales chalybeiformis* with *Gyrophora cirrhosa*, *Lecanora frustulosa* and *L. melanophthalma*. The vertical surfaces are occupied by the *G. cylindrica-Cetraria noermoerica* association.

The nitrophilous bark communities of lichens, mosses and algae have been carefully examined by OCHSNER (1925) and FREY (1928) (See BRAUN-BLANQUET 1932). OCHSNER distinguishes two markedly nitrophilous bark associations; (i) the light loving *Physcietum* and (ii) the *Parmelietum acetabulae* comprising several sub-associations, which prefers older trees with furrowed barks.

The range of the nitrate factor has been experimentally proved at Rothamstead where with the application of nitrogenous manure and especially with nitrates, grasses preponderated and expelled the leguminous plants, whereas the presence of potassic salts favoured the latter. Experimental manuring of high moors, according to Weber as quoted by WARMING (1925), has led to similar results.

It can be seen from the review of literature, that practically no work on the subject is done in India except by MISRA (1944) as far as the authors are aware.

DISCUSSION.

From the definition of WARMING it becomes clear that the plants in question thrive best in soils rich in nitrates and other plants cannot grow in such habitats. Thus he confirms to the first view that nitrates are necessary for nitrophilous plants.

BAUER's definition merely implies that the plants accumulate nitrates and they are capable of enduring high concentrations of the salt. He grades plants according to the decreasing amounts of nitrates stored in their tissues and also according to their

capacity of "enduring" nitrates in high concentrations. From his work it is clear that nothing more than mere capacity to endure and accumulate nitrates is the basis of nitrophily. Hence his definition is nearer the second view namely the capacity to tolerate high nitrate-concentration.

CARSTEN OLSEN also asserts the capacity of the nitrate plants to absorb and assimilate nitrates. He further adds that when not assimilated, they are accumulated. The very fact that they greedily absorb nitrates and assimilate them, shows that nitrates are necessary to their well being and thus his definition of nitrophily is nearer the first view i.e. that nitrates are necessary.

The capacity of nitrophilous plants to accumulate nitrates in their cell-sap has also been mentioned by both BRAUN-BLANQUET as well as HESSELMAN, though none of them mention whether the role of nitrates is active or passive.

The application of nitrogenous manure giving rise to a preponderance of grasses and the expulsion of the leguminous plants as mentioned earlier, illustrates the dynamic role of the nitrate factor and shows how nitrophilous communities can be formed in nature.

From the definitions advanced by various authors, we find that not one of them definitely and explicitly mentions whether a high concentration of nitrates is absolutely necessary to a nitrophilous plant or whether it can merely endure nitrates in high concentrations. The confusion is due to the fact that none of them have realised that there are degrees in nitrophily. The whole range of nitrophily stretches from plants absolutely restricted by nitrates to ones indifferent to them and yet occurring on such habitats. The plants can thus be classified in the same way as UNGER classified them with regard to chalk and silica, namely (i) indifferent to such soils (ii) partial to such soils and (iii) restricted to such soils. Thus whether nitrates are necessary to a nitrophilous plant or otherwise, depends upon what we mean by a nitrophilous plant and upon its place in the classification.

Nitrophilous plants as understood from the plant indicator point of view are only those which *indicate and characterise the habitat*, but we have just seen that there are other plants in the lower grades of nitrophily which cannot indicate the habitat since they can also occur on other soils (Nitrate-normal or nitrate-low). This specific property of the characteristic species to grow only in high-nitrate habitats is important, since if a high-concentration were not necessary to the nitrophilous plant and its presence was merely due to its capacity to endure nitrates, the plant as such could have no indicator value, for then it is reasonable to suppose that it would occur equally often in other habitats as well. But this is not the case as can be seen from the reports of various workers referred to previously where nitrophilous communities indicating high-nitrate habitats are mentioned (CARSTEN OLSEN, SERNANDER, FREY, GAMS and MOTYKA).

THE PHYSIOLOGY OF NITROPHILY.

The problem of nitrophily shows a number of anomalies when examined from the physiological point of view as from the ecological.*) It has been known from the times of KNOPS and SACHS that plants could be made to grow without soil provided

* Recently much interest seems to be shown in the problem of nitrogen metabolism as evinced from the following three articles which we cite merely as references since they hardly touch the aspect of the problem that we have dealt with:

1. MCKEE, H. S., 1949 — Review of recent work on Nitrogen Metabolism. *New Phytologist*, **48**, 1.
2. STREET, H. E., 1949 — Experimental methods available for the Study of the nitrogen metabolism of plants. A review of some recent advances. *Ibid.* **48**, 1.
3. PEARSALL, W. H., 1949 — Nitrogen metabolism in plants. *Endeavour*, **8**, 31.

that they could get what they needed in solution form. It has also been known since very early times that plants cannot survive a solution where the nutrient salts are not properly balanced. KAHLENBURG and TRUE as far back as 1896 have expressed the toxicity of different mineral salts in respect to Wheat by a number called equivalent toxicity which represents the minimum quantity in grams which when dissolved in 100 ccs of water will cause the death of the plant. Thus according to COUPIN (see RABER 1937) equivalent toxicity of Ammonium nitrate = 3.9, that of Potassium nitrate = 3.0.

The nutrient salts used in hydroponic solutions balance the toxic effect of one another by virtue of their property of antagonism. The main question at issue is, therefore, how the nitrates in a nitrophilous habitat do not become toxic to the plants which thrive upon it and also whether the excess so absorbed is balanced internally by the plant.

It would also be logical to ask whether the absorption of nitrates is at the cost of another element. This has theoretical support since a plant cannot absorb an excess of one element without releasing another in equivalent quantity e.g. GEORGE HOFFER proved that maize accumulated a large excess of Iron when the soil was deficient in Potassium. This excess blocked up the xylem and ultimately laid the plant open to the attack of root parasitic fungi. JOHNSTON and HOAGLAND (1929) found that a low supply of Potassium in the soil caused an increased absorption of Calcium, Magnesium and Phosphorus, an observation substantiated by the work of COLBY (1933) on French prune trees and PHILLIPS, SMITH and DEARBORN (1934) on tomatoes. A colleague working on the die-back disease of citrus in our own laboratory, has come to the conclusion that deficiency of B, Cu and Zn causes the citrus plants to accumulate calcium oxalate in quantities far greater than in normal plants. Or the accumulation of nitrates in the cell sap of plants may be due to a faulty synthesis of proteins. One of the factors which favours protein synthesis is the presence of Phosphorus. It was found by KRAYBILL (1930) that nitrates are not reduced by tomato plants in the absence of phosphates. ECKERSON (1929) (1931) noted that depletion of phosphates in the tomato plants caused a rapid decrease of the reducing enzyme and an accumulation of nitrates.

The role of Potassium as a nitrate reducer has been worked out by NIGHTINGALE, SCHERMERHORN and ROBBINS (1930). They consider that it is apparently essential, either directly or indirectly for the initial stages in nitrate reduction in plants and probably in the synthesis of Proteins in meristematic tissue.

Turning to the soil we find that the high-nitrate habitat depends quite a lot upon the deposit of organic matter in the soil and also upon its capacity to ammonify and nitrify it. It has recently been demonstrated in our laboratory that soil samples collected from under nitrophilous plants always show a very high capacity for nitrification.

Plants in general absorb nitrogen either in the cationic form as ammonia or in the anionic form as nitrates, depending mainly upon the pH. The observation that plants absorb nitrogen in the form of ammonium salts more rapidly from the nutrient medium at a relatively high pH, while nitrogen in the form of nitrates is absorbed more rapidly at a lower pH value has been reported by ADDOMS and MOUNCE (1932) for young Cranberry plants, by TIEDJENS and BLAKE (1932) for Apple trees, by CLARK (1933) on Strawberry plants, by DAVIDSON and SHIVE (1934) for Peach trees and by TIEDJENS (1934) for Tomato, Cotton and the seedlings of several varieties of Apple. According to ARRINGTON and SHIVE (1935), in tomato plants the rate of absorption of Cation N from a medium with pH 7 was more than three times as high as the rate at pH 4. For anion N the rate of absorption was somewhat greater at pH 4 than at pH 7. CONRAD (1934) considered that a plant absorbs the nitrate ion more rapidly from a more acid medium because it can secure more

easily from it the requisite H ions to accompany the nitrate ions into the plant. The same reasoning is applied for the absorption of ammonia ions from the more alkaline solutions. In so far as the authors are aware, all these observations have been made from the work on cultivated plants. If the same is true for wild plants also, then the definition of CARPENTER (1938) of nitrophilous plants as Alkali lovers, becomes even more difficult to understand.

The difference in the absorption rate of the two ions of a salt makes a great change in the pH value of the habitat. If bases are left in the soil it will tend to become alkaline as when the nitrate ion is used, while if acid groups are left the soil becomes more acid (as when the ammonium ion is used). In general the plants tend to absorb ions in such a way that the result is to bring the soil reaction nearer to neutrality. Thus if a plant were grown in an acid soil the soil reaction in the vicinity of the plant could be made more nearly neutral by absorbing the acid radicals from the soil or by actively excreting alkaline substances. JACOBSON found that hundred days old wheat plants changed the reaction of the culture solution in 12 hours from pH 3.9 to 6.3, due possibly to greater absorption of the nitrate ion. Plants in general tend to bring the reaction of the substratum to pH 5.0—7.0 when placed in solutions more acid or alkaline than these (see RABER, 1937).

This 'buffering' effect is coordinated with the process of nitrification. Thus HESSELMAN (1926) from his work on Swedish forest soils has proved that ammonification is at its optimum at pH 4.5 - whereas nitrification is at its maximum at pH 6 or more. Therefore we assume that when the plant absorbs the ammonium ion it automatically brings the soil within a pH range where more ammonia can be formed, namely round about pH 4.5—5. Conversely when the plant absorbs more of anionic nitrogen the soil gradually turns alkaline thus reaching a pH value at which more nitrates can be formed.

A study of the effect of soluble salts on ammonification and nitrification, according to LIPMAN, GREAVES and SACKETT and COLLINS shows that soluble salts are generally speaking harmful though at low concentrations they produce a stimulating effect. In general chlorides are the most toxic, while nitrates, sulphates and carbonates are successively less toxic to the ammonifiers but the order is almost reversed for the nitrifiers i.e. in the following order of toxicity: Carbonates > Nitrates > Sulphates > Chlorides. The fact that the ammonia producers can tolerate soluble salts better than the nitrifiers is fortunate, since if the reverse were to be the case, ammonification would stop after a time and consequently there would be no nitrates formed.

Amongst these, the reactions to chlorides, carbonates and nitrates are noteworthy. The toxicity due to chlorides is important if considered from the point of view of Saline Soils. This has been experimentally verified in our laboratory, where from the results obtained so far it has been noted that members of the halophilous *Chenopodiaceae* do not show any nitrates in their tissue, in spite of the fact that it has been put down by WARMING (1925) as a nitrophilous family. It has also been noted from our experiments on the nitrifying capacity of mangrove soils that such soils show very poor capacity for nitrification. It is also fortunate that the carbonates are not so very toxic to ammonifiers as they are to nitrifiers since their union with ammonia to form ammonium carbonate is an essential step in nitrification. The fact that nitrates are toxic both to ammonifiers and to nitrifiers illustrates how in nature after a certain quantity of nitrates are produced the whole process slows down until the excess is lessened, whereupon due to its stimulating effect at low concentrations, the process is vigorously resumed. It thus becomes clear that the producing mechanism is very intricately balanced and illustrates the economy of nature. It shows how overproduction of nitrates is checked and how due to stimulation at low concentrations a certain level of nitrates is maintained as long as there is a sufficient supply of organic matter in the soil.

Though the producing agencies are so well regulated, the same cannot be said of the agencies which destroy or remove the nitrates formed from the soil as said earlier. They consist of many unpredictable factors like bacteria, plants and rain. The soil organisms take up nitrates, phosphates and other minerals and they are able to get them more readily than the plants. It is because of this that partial sterilization of the soil leads to increased fertility due to the fact that organisms surviving partial sterilization produce more ammonia and nitrates than the original population. Rainfall also causes great losses in nitrate nitrogen since they are freely soluble in water and hence are washed away. Thus the nitrate content of a particular habitat falls down very much after a shower. It thus becomes clear that it is futile to look for the evidence of nitrification in the soil. Various workers like RUSSEL (1915) and OLSEN (1921) have expressed the opinion that the evidence of the presence or the absence of nitrates in the soil is illusory. RUSSEL mentions the fact that the amount of nitrate in the soil at any time cannot be taken as the measure of the rate of nitrate production, the quantity that is actually required. He further adds that the amount of nitrate in the soil at any time only represents the balance of gain over losses. The same author has mentioned elsewhere (1932), that the fluctuations in the nitrate-content are very considerable and are due to the tug-of-war between the producing agencies and the removing agencies in the soil. He then adds that since ammonification is the slowest process it sets a limit to nitrification as a whole and agrees with GAINEY, that a measure of the speed of nitrate formation does not measure the rate of nitrification but of ammonification.

BRAUN-BLANQUET (1922) has also mentioned that the amount of nitrate present in the soil at any time gives no indication of the nitrate supply of a plant community. He also refers to the removal of nitrates from the soil by rain and plants and quotes the work of OLSEN (1925) who showed that the soil of flat moor communities with *Carices*, *Molinia* and *Deschampsia* and others which did not show a trace of nitrate at the first estimation, showed 22 to 25 mg/litre of soil after 25 days. CARSTEN OLSEN in his "Ecology of *Urtica dioica*" mentions that since nitrates are greedily taken up from the soil by the plants and being easily soluble are leached out by rain, it makes the quantity of nitrates very variable at different times and estimations of nitrate content in soil samples, cannot therefore be used.

A further draw back in soil analysis is provided by the fact that the soil continues to nitrify even after it is sampled, which produces a large error, especially if a long time elapses between the sampling of the soil and its subsequent analysis. Thus PIPER, in his "Soil and Plant Analysis" (1944) has recommended heating the soil sample as soon as possible to 60° C for 2 hours to kill all the organisms taking part in nitrification. A similar procedure is also recommended by KNOWLES and WATKINS, in their book "A Practical Course in Agricultural Chemistry" (1937) where the sample taken direct from the field is dried up at once at 55° C in order to prevent nitrification.

Apart from the evidence cited above, the lack of correlation between the nitrates in the soil and those in the plant has been confirmed experimentally by us in our laboratory. It thus becomes clear that the nitrate-high habitat is exceedingly difficult to define since nitrification is such a complex process governed by a number of factors and subject to unpredictable losses.

To conclude, therefore, the problem of nitrophily resolves itself into two branches of study viz. autecology and synecology of nitrophilous plants.

A complete autoecological study of the problem should seek to answer the following questions:

- 1) Why is it that nitrates in such large quantities are not toxic to the nitrophiles when in similar concentrations they are quite lethal to other plants?
- 2) Do nitrophiles absorb an excess of nitrates because of any deficiency in the

soil of either K_2 or PO_4 etc. or is it because they really thrive upon them, or is the excess of nitrate nitrogen in their tissue due to a faulty synthesis of Proteins?

3) Do nitrophiles absorb any nitrogen in the form of ammonia and if so, what is the ratio of the nitrogen absorbed as anion and as cation?

On this ratio will depend the important factor of the pH of the plants. Absorbing an excess of the acid nitrate radical must tend to make the cell sap acid beyond normal. How does the plant then keep its pH balance?

Synecology amounts to a survey of the nitrophilous plant associations of the province. It started first with laying down a criterion for the determination of nitrophily and ended with a synthesis between the sociological method of BRAUN-BLANQUET and the bio-chemical method devised by us. This way we were able for the first time to grade the plants according to their degree of nitrophily which was accomplished by means of a new formula proposed by us. This has been illustrated by a diagram and it was later incorporated in the study of the Calciphilous plants by a colleague in our laboratory. The criteria followed and the methods used, will be discussed later.

It can thus be seen that the problem of nitrophily is not just one problem but many problems; it does not depend upon one aspect of the vegetation but upon many aspects, several of which are now being tackled in our laboratory.

We however believe that upon this problem depends at least in part the entire problem of the effect of the soil on the vegetation in general, and the effect of the vegetation on the soil which thus intimately touches the entire domain of phytosociology.

THE CRITERION OF NITROPHILY.

It is clear that nitrophilous plants are those plants which thrive in a habitat from which they can derive the maximum of nitrogen in the form of nitrates and that they show a consistent presence of a large amount of the salt in their tissues. That other plants can grow but very feebly on such habitats indicates the dynamic role of the soil factor.

The fact that nitrophilous plants contain a large quantity of nitrates also indirectly points to the soil since the nitrates in the plant could have come only from the soil. In plants, other than nitrophiles an accumulation of nitrates in the tissues points either to faulty protein synthesis or to a deficiency of some vital element in the soil. Both these conditions are so to say departures from the normal and are likely to occur in any species where the conditions for soil nutrition are abnormal. Moreover such plants can have no possible phyto-sociological value either as characteristic species or as dominants in any association.

Nitrophilous plants on the other hand, accumulate nitrates because it is in their nature to do so. They cannot grow well enough in any other habitat, which does not yield enough of nitrogen as nitrate. As regards the role of the nitrate factor in their metabolism it is too early to surmise, just as it is with regard to an accumulation of carotene in carrot root or the accumulation of ascorbic acid in plants like *Citrus*, *Tropeolum* etc. According to MCKEE (loc. cit.) accumulation of a given metabolite is due to a lack of balance between its formation and utilization and that such an accumulation suggests a deranged metabolism.

But the fact, however, remains that only a few definite plant species can grow in such a habitat at the cost of all the rest. This lends a phytosociological colour to the whole problem and differentiates it from mere physiology. That such plants belong only to certain specific families is a statement first made by WARMING (1925) and seems to have been borne out to some extent by our own work. This gives further support to our contention that these plants with a deranged metabolism and a tendency

to withstand or even thrive upon lethal quantities of nitrate nitrogen form a close knit community in themselves, and thus have a profound ecological significance.

The criteria of nitrophily are two, viz. (i) nitrates in the soil and (ii) in the plant.

As has been said before the nitrates in the soil can give no indication of nitrophily.

It is however possible to look for the evidence of nitrophily in the rate of nitrification of the soil. A better method of doing so has now been evolved in our laboratory and from the results obtained so far, it seems that a regular correlation exists between nitrophily and nitrification, and that the rate of nitrification will provide an important factor in judging nitrophily. But at the moment, the evidence obtained from the cell-sap of plants seems more reliable.

The soil, therefore, is a very dynamic factor and there are yet many aspects of it which are exceedingly difficult to estimate and standardise. Thus the only reliable evidence is the one which can be obtained from the nitrate analysis of the cell-sap of plants. According to BRAUN-BLANQUET (1932), this idea was first mentioned by HESSELMAN (1917) and RAUNKIER (1926) and depends upon the principle that all the nitrates that a plant contains can only come from the soil. Thus a number of plants showing a high-nitrate concentration of their cell-sap give a better clue to a high-nitrate habitat than any other method used today. BAUER, CARSTEN OLSEN and BRAUN-BLANQUET have used the nitrate content of the cell-sap as an indication of nitrophily, as can be seen from the literature cited above, but whether the mere presence of nitrates in the cell-sap could provide a complete clue to the complex and varying degrees of nitrophily is doubtful.

FORMULA PROPOSED.

Nitrophily does not merely depend upon the capacity of the plant to accumulate nitrates but also to a large extent upon its indicator value, in other words upon the frequency of its presence in high-nitrate habitats.

From the literature cited, the following facts are noteworthy:

(i) Nitrophilous plants are characterised by their capacity to accumulate nitrates in their tissues.

(ii) BAUER has graded plants according to their capacity to endure nitrates in high concentrations. He has also graded them according to the quantities of nitrates stored in their tissues. CARSTEN OLSEN has also used the quantity of nitrates as an indication of nitrophily. Thus nitrophily can also be defined from the quantity of nitrates accumulated by the plants.

(iii) Nitrophilous plants have indicator value, i.e. a high-nitrate habitat can be made out from the rich growth of plants characterized by their having large quantities of nitrates accumulated in their tissues.

Thus nitrophily is governed not by one factor but by three measurable factors, namely (a) Frequency (b) Constancy of nitrates and (c) Average nitrate-content. We shall deal with each factor separately and illustrate our hypothesis with local examples.

a. Frequency.

This factor measures the frequency of a particular plant in all the high-nitrate localities examined. It is measured by the number of times the particular plant is present divided by the total number of high-nitrate places investigated. The number so obtained yields a relative value for the frequency of the plant in the high-nitrate habitat e.g. *Amaranthus spinosus* = 0.374, *Portulaca oleracea* = 0.1, *Euphorbia pilulifera* = 0.075, *Solanum xanthocarpum* = 0.1.

b. Constancy of Nitrates.

It is measured as the number of times the plant gives the positive nitrate test out of the total number of times it is analysed. Thus, *Amaranthus spinosus* = 0.588, *Portulaca oleracea* = 1, *Euphorbia pilulifera* = 0.5, *Solanum xanthocarpum* = 1.

c. Average Nitrate-Content.

Without this factor, no estimate would be correct, since the concentration of nitrates in the cell-sap, gives a quantitative value of the nitrate accumulating power of the plant viz. - *Amaranthus spinosus* = 165 p.p.m., *Portulaca oleracea* = 155 p.p.m., *Euphorbia pilulifera* = 56 p.p.m., *Solanum xanthocarpum* = 311 p.p.m.

It can be seen from the above examples that each factor if considered separately would yield a different order of nitrophily. It stands to reason, therefore, that no one or two factors by themselves could give a correct idea of nitrophily as can be seen from the case of *Solanum xanthocarpum*, a plant which though rarely found in dirty places (and hence with very little indicator value) could easily be taken as very nitrophilous if considered solely on grounds of its average nitrate-content.

The exact range of each of these factors cannot as yet be put down mathematically until much more work is done on the subject. We cannot, for instance, formulate that a certain factor is say twice or three times as important as the other two. Hence all three factors can provisionally be taken as equal in magnitude and importance till more definite data are obtained. Thus the degree of nitrophily N, could be formulated as $N = A \times B \times C$, where A, B, and C are frequency, constancy of nitrates, and the average nitrate-content respectively.

Only on considering the product of the three factors can the plants be graded fairly in a descending order of nitrophily. Thus:

$$\begin{aligned} \text{For } & \textit{Amaranthus spinosus}, \\ & N = 0.374 \times 0.858 \times 165 = 52.8 \\ \text{For } & \textit{Portulaca oleracea}, \\ & N = 0.1 \times 1 \times 155 = 15.5 \\ \text{For } & \textit{Euphorbia pilulifera}, \\ & N = 0.075 \times 0.5 \times 56 = 2.1 \\ \text{For } & \textit{Solanum xanthocarpum}, \\ & N = 0.1 \times 1 \times 311 = 31.1. \end{aligned}$$

Hence according to the formula, the order of nitrophily of the above plants would be as follows:

- (1) *Amaranthus spinosus*,
- (2) *Solanum xanthocarpum*,
- (3) *Portulaca oleracea*,
- (4) *Euphorbia pilulifera*.

ERRORS AND CORRECTIONS.

The sources of error for this formula are not many, provided a large number of relevés are investigated. They could be briefly termed as due to:

a. Irregularity of Seasons.

Due to the failure of a particular seasonal factor, certain plants may not appear upon the habitat in their usual proportion and quota, as happened in Bombay in the year 1946-47, when due to the failure of the cold season, *Solanum xanthocarpum* did not appear in its usual quantities, and thus it would not be fair to compare its

nitrophily to that of plants like *Amaranthus spinosus* or *Portulaca oleracea* which appeared in their usual proportions and abundance.

The error though formidable could be easily avoided by considering nitrophily not merely on investigations of a year or two but of many years, as far as possible with normal seasons.

b. Seasonal Distribution of Plants.

At first sight it does not also seem correct to compare the nitrophily of plants which only grow in a particular season, like *Solanum xanthocarpum* with plants like *Amaranthus spinosus* which grows practically throughout the year. This could be obviated by considering the seasonal nitrophily of a habitat. Thus the habitat could have a different set of nitrophilous plants for different seasons.

c. Relative Importance of Factors.

As mentioned before, the three factors may not be equal in importance and magnitude and their exact mathematical relationship is debatable. This error if at all, would be universal and hence could be overlooked until further data are available.

Thus, on the whole, in spite of the various errors mentioned the formula for the time being can be provisionally accepted. It can also be used for the calciphilous plants as well as for any plant community which is regulated by a particular chemical factor, with suitable modifications. Its importance does not merely lie in the fact that it allows us to measure nitrophily but that it can be used for the measurement of various other aspects of plant sociology governed by particular chemical or physical factors. We feel that provisionally it will go a long way towards giving a clear and concise idea of nitrophily until it has been suitably modified in the light of further research, or replaced by a better one.

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BIBLIOGRAPHY.

- ADDOMS R. M. & F. C. MOUNCE, 1932 — Further notes on nutrient requirements and histology of the cranberry, with special reference to the sources of nitrogen. *Plant Physiol.* **7** 643—656.
- ARRINGTON L. B. & J. W. SHIVE, 1935 — Rates of absorption of Ammonium & Nitrate nitrogen from culture solutions by ten-day old tomato seedlings at two pH levels. *Soil Sci.* **39**, 431—435.
- BAUER, J., 1938 — Beiträge zur Physiologie der Ruderalpflanzen. (An ecological study of the flora of the city dump of Leipzig-Mockern) *Planta* **28**, 3, 383—428.
- BRAUN-BLANQUET, J., — *Plant Sociology*. McGraw-Hill Book Co. New York, 1932.
- CARPENTER, J. R., — *An Ecological Glossary*. Kegan Paul, Trench, Trubner & Co., Ltd. London, 1938.
- CLARK, J. H., 1933 — Reaction of the nutrient medium as affecting growth of strawberry plants. *Proc. Am. Soc. Hort. Sci.* **30**, 283—287.
- COLBY, H. L., 1933 — Effect of starvation on distribution of mineral nutrients in french prune trees grown in solution cultures. *Plant Physiol.* **8**, 357—394.
- CONRAD, J. P., 1934 — Physiological acidity and alkalinity of inorganic nitrogenous compounds in solution cultures. *J. Amer. Soc. Agron.* **26**, 364—372.
- CUTLER, D. W. & B. K. MUKHERJI, 1931 — Nitrate formation by Soil bacteria. *Proc. Roy. Soc. B.* **108**, 389.
- DAVIDSON, O. W. & J. W. SHIVE, 1934 — The influence of the hydrogen-ion concentration of the culture solution upon the absorption and assimilation of nitrate and ammonium nitrogen by peach trees grown in sand cultures. *Soil Sci.* **37**, 357—386.

- ECKERSON, S. H., 1929 — Influence of phosphorus deficiency on metabolism of the tomato plants. *Abst. Am. J. Bot.* **16**, 852.
- ECKERSON, S. H., 1931 — Influence of phosphorus deficiency on metabolism of the tomato (*Lycopersicon esculentum*) *Contr. Boyce Thompson Inst.* **3**, 197—217.
- HESSELMAN, H., 1917 — Studier över salpeterbildingen i naturliga jordmaner. *Medd. Fran Stat. Skogsförsöksanstalt.* 13—14.
- JACKSON, B. D., 1928 — *A Glossary of Botanic Terms.* Duckworth, London.
- JOHNSTON, E. S. & D. R. HOAGLAND, 1929 — Minimum potassium level required by tomato plants grown in the water cultures. *Soil Sci.* **27**, 89—109.
- KRAYBILL, H. R., 1930 — Plant metabolism studies as an aid in determining fertilizer requirements. I. *Ind. & Eng. Chem.* **22**, 275—276.
- MILLER, E. C., 1938 — *Plant Physiology.* McGraw-Hill Book Co., Inc. N.Y. & London.
- MISRA, R. D., 1944 — *The soil-complex in Ecology.* (Presidential address delivered to the Benares Centre of the Indian Ecological Society. 9th Dec. 1944).
- NIGHTINGALE, G. T., L. G. SCHERMERHORN & W. R. ROBBINS, 1930 — Some effects of potassium deficiency on the histological structure & nitrogenous and carbohydrate constituents of plants. *N. J. Sta. Bull.* **499**.
- OLSEN, C., 1921 — *Ecology of Urtica dioica.* *J. Ecol.* **9**, 1.
- PHILLIPS, T. G., T. O. SMITH & R. B. DEARBORN, 1934 — The effect of potassium deficiency in the composition of the tomato plant. *N. H. Sta. Tech. Bull.* **59**.
- RABER, O., 1937 — *Principles of Plant Physiology.* The MacMillan Co., New York.
- RUSSEL, E. J., 1915 — The nature and amount of fluctuations in nitrate content of arable soils. *J. Ecol.* **3**, 233.
- RUSSEL, E. J., 1932 — *Soil conditions and Plant growth.* Longmans Green & Co., 6th edition, London.
- TIEDJENS, V. A., 1934 — Factors affecting assimilation of ammonium and nitrate nitrogen, particularly in tomato and apple. *Plant Physiol.* **9**, 31—57.
- TIEDJENS, V. A. & M. A. BLAKE, 1932 — Factors affecting the use of nitrate and ammonium nitrogen by apple trees. *N. J. Sta. Bull.* **547**.
- UNGER, F., 1836 — *Ueber den Einfluss des Bodens auf die Vertheilung der Gewächse,* Vienna.
- WARMING, H., 1925 — *Oecology of Plants.* Oxford University Press, London.