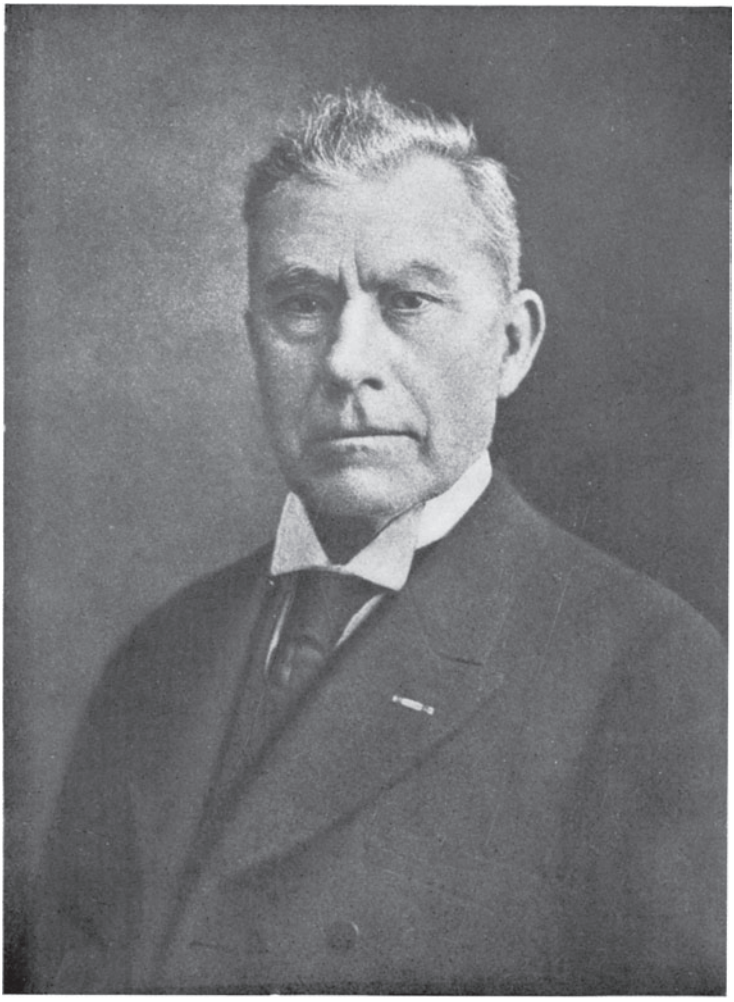
PURE CULTURES OF ALGAE



M. W. BEIJERINCK

PURE CULTURES OF ALGAE

Their Preparation & Maintenance

BY

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A NOTE ON THE FRONTISPIECE

In selecting M. W. Beijerinck's *portrait as the frontispiece of this book, the author has been guided not only by the consideration that* Beijerinck *was the first to apply the technique of bacteriology in the preparation of pure cultures of algae, but that, in company with* Pasteur *and* Winogradsky, Beijerinck *also laid the foundations of General Microbiology.*

The present author has, throughout his career, been greatly influenced by Beijerinck's *practice of securing enrichment of specially adapted ecological types by a consideration of their biological properties, thus achieving what he called 'natural pure cultures'. Such cultures have proved very helpful in elucidating the physiological interrelation between algae and other micro-organisms, without a knowledge of which we cannot hope to understand their conditions of life or proceed methodically to obtain pure cultures, which must form the basis of exact biochemical research and of the investigation of the physiology of development.*

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FOREWORD

In many branches of biological science progress is dependent on the gradual improvement of the methods of investigation. A methodical study of the enormous diversity of simple forms of life, in part differing among one another only in slight and rather elusive particulars, is only possible if extensive and homogeneous material becomes available. Attempts to obtain such material by artificial means, based on the technique developed for bacteria and later for fungi, were therefore instituted already in the last century, and Pringsheim rightly hails Beijerinck as a pioneer in this direction. Slightly later a vigorous development of the culture technique followed at Geneva and, under the leadership of Robert Chodat, numerous green algae were grown in this way. The remarkable polymorphism, and the bizarre forms often appearing in his and his collaborator's cultures, however, gave rise to an element of scepticism as regards the value of such cultures, which found expression in various ways.*

Meanwhile, however, the technique of pure culture was being investigated by the author of this book from a new angle. Recognizing the diversity of ecological factors to which the organisms concerned are subjected, Pringsheim developed a more elastic method of approach and endeavoured to mould his culture technique to fit the needs of the individual alga or flagellate. How great a degree of success has thus been achieved will be evident to anyone who has made use of the numerous cultures he has now at his disposal. These cultures are healthy populations composed of single organisms, and in general display none of that extraordinary

* Cf. e.g. Fritsch, Presidential Address, Brit. Assoc., Leeds, 1927, Sect. K, p. 15.

variability which made one feel dubious as to the value of the cultures grown by the Geneva school.

Moreover, Pringsheim has recognized the fundamental fact that, unless required for special purposes, in particular for physiological investigations, it is unnecessary to have bacteria-free cultures. His soil- and water-culture method, offering as it does scope for unlimited variation in the cultural conditions, marks in my opinion one of the biggest contributions he has made to the methodology of the culture of lower organisms. Since in nature Algae and Flagellata are always associated with bacteria, it is probable that the latter may often play an essential role in relation to the life processes of the former. The absence of such cooperation in bacteria-free cultures may often be one of the prime causes of failure to secure them, although it is clear that with many lower organisms growth in the absence of bacteria can be successfully achieved.

The author himself enumerates towards the end of this book the many diverse purposes for which unialgal or bacteria-free cultures can be utilized. It cannot be doubted that genetically homogeneous material of the simpler forms of life will be used to an increasing extent in the study of fundamental physiological principles. At the same time the perfection of methods of culture will render feasible a more direct approach to some of the problems of fresh-water biology than has hitherto been possible. A reassembly of mixed populations, both of microscopic plants and of animals grown individually in pure culture, should render possible a clearer elucidation of the problems of competition under standardized conditions and of the mutual interreactions of the components of a population upon one another. This may well indeed constitute a major step in the elucidation of the first links in the food chain.

The possibility of rearing cultures from a large diversity of lower organisms entails the risk that this may come to be

an end in itself, and that many of the forms involved become known to science only in this condition, without definite information as to their mode of occurrence in nature.* The pursuit of inquiry along these lines will often provide data of appreciable morphological or physiological interest and may widen our taxonomic outlook, but, until the role and mode of occurrence of such forms in nature have been established, the information gleaned from their culture is largely academic and is of practically no value to the ecologist. Pringsheim therefore rightly emphasizes the importance of a preliminary study of all organisms taken into culture, both with a view to ascertaining their conditions of existence and to establishing their mode of occurrence in nature.

Any scientific method depends for its success on attention to a multitude of detail and requires the exercise of considerable patience on the part of the investigator until the requisite manipulative skill has been acquired. It is impossible to overestimate the importance of data supplied as a result of experience accumulated over many years by an expert like Pringsheim, and this volume should go far in the direction of initiating the novice into the methods of pure culture and of fostering interest in this important technique. If it achieves the success which it deserves, it may well open up a new era in the intensive investigation of the many aspects of lower organisms that claim the immediate attention of biologists. In many ways the study of this branch of microbiology is of outstanding economic importance, quite apart from its fundamental interest in exposing the characteristics and modes of life of lower forms of plant and animal organization.

F. E. FRITSCH

Cambridge
June 1944

* Cf. for instance the large number of new Heterokontae described by Pascher in vol. XI (1937) of Rabenhorst's *Kryptogamenflora*.

PREFACE

Since I described the methods of working with cultures of algae used in my laboratory at Prague (1926 *a*) there has been no really useful publication in this field of biology until Bold (1942) gave a review based on literature* and supplemented it with valuable comments. In the meantime, however, the technique has been further developed, so that it is now easier to prepare pure cultures, and the method has been extended to embrace more species.

On the other hand, it is becoming increasingly obvious that bacteria-free cultures are needed, not only for experiments on nutrition, but also for certain problems of ecology, hydrobiology and fishery which cannot be effectively approached without such cultures.† Many problems relating to propagation, heredity and genetics cannot, moreover, be solved without the help of at least unialgal cultures. Even the original aim of algal study, namely, that of describing species, cannot, as we have been forced to acknowledge, always be attained without cultures starting from single cells. For these reasons almost all workers on algae will in future be obliged to use cultural methods.

My object is to show that it is by no means difficult to acquire the necessary skill. With some patience and zeal, valuable results can be obtained after a relatively short period of training in this attractive field of biology.

* Bold's paper gives an almost complete list of relevant references.

† I do not wish to give the impression that I regard pure cultures as indispensable for ecological work in the field. Such over-emphasis could only be harmful to the adoption of the technique of pure cultures. Simple cultural methods will, however, prove very helpful to workers in this field, and investigations based on pure cultures will afford a foundation for future floristic and applied work on algae.

By publishing an account of the practical conclusions reached during many years of experimentation with algae, I hope to draw the attention of biologists to the great possibilities opened up by the culture of these organisms. Existing circumstances have not for some time afforded me the opportunity of training junior research workers, which would have been a much more effective method of imparting instruction than a written account can ever be. If fortune favours me, I hope to be able to act in this capacity again. The progress of science depends not only on research, but also on tradition. Just as strains maintained in my culture collection have been available to everyone, I would like to give all who are interested the opportunity of avoiding my mistakes and of profiting by my experience in this field of biology. I still hope to find the means for the establishment of a small station, devoted to research on algae.

For those who question the value of such an institution the following considerations are relevant. Much time and labour have been wasted by the failure to define and recognize species of algae and flagellates. Much physiological research has been invalidated by the use of impure or unidentified algae or could not be undertaken for lack of suitable material. Many morphological investigations have been interrupted or abandoned because living material was no longer available. Many theoretical problems are best solved with the help of these small organisms. Test objects suitable for medical and other purposes could probably be found among the cultivated species. Little is known about vitamin production, about the sources of food for animals of vital interest to mankind, about the origin of organic substances in sea and fresh water, about the relation of soil algae to fertility, and so on. These problems are likely to be solved with the help of pure or combined mass cultures and their biological and chemical investigation. Nobody should therefore overlook the probability that we

are at the threshold of a scientific edifice which must be built sooner or later, comprising the specific ecology and physiology of organisms of which only morphological data are as yet known.

Many of the experiments and observations which form the basis of the experience detailed in the following pages are the fruits of the years spent in England after I had been deprived of my working place in Prague. I find it difficult to express my indebtedness to those who made it possible for me to continue my researches, but special mention should be made of the Society for the Protection of Science and Learning, the Royal Society of London, the British Association for the Advancement of Science, and the University of London, who assisted me with grants. Many thanks are due to Queen Mary College, University of London, and especially to the head of its Department of Botany, Prof. F. E. Fritsch, who did all in his power to improve the conditions of work, and who encouraged me much by his friendly interest; as well as to the staff of the Botany School in the University of Cambridge, where I carried on my work after Queen Mary College was evacuated from London. Without the help of my wife, my only assistant, I could not have preserved the culture collection on which my investigations are based.

E. G. P.