

Conceptual Context with Some Expert Comments on Identification Avian Malaria Parasites: A Learner's Experience

NC Nandi*

Social Environmental and Biological association Kolkata, India

*Corresponding Author: NC Nandi, Social Environmental and Biological association Kolkata, India.

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Introduction

Initially, earlier researchers were inclined to the idea of 'one host – one parasite' concept and often describing new species from a single blood smear without adequate descriptions. Comparison of collected materials in those days was impossible with that from other regions even though identification of animals, especially parasites is a very vital issue in the spheres of Veterinary Parasitology and Malariaology. It is a fact that medication and/or management actions are mainly based on accurate identification of the parasites. Species level diagnosis and symptoms may lead to proper treatment as there are species specific variances in disease related clinical responses. As such, this communication is intended to help facilitate identification of malarial parasites and its closely allied relatives primarily based on blood smears so that prompt and proper treatment/management of wild and pet birds is possible. This is also because of the problems of species identification faced by this author while doing his doctoral dissertation. The responses to my queries from different experts are also quoted here, under expert comments (Please vide infra) to share it as a learner's experience.

Conceptual Context

Taxonomically members of the family Plasmodiidae and the genus *Plasmodium* are popularly known as malarial parasites. Herein, representative species of the genus *Haemoproteus* are also considered close relative of malarial parasites for producing haemozoin pigment granules and also for earlier inclusion of these two genera in the same family Plasmodiidae, Now, species of the genus *Haemoproteus* embrace a separate family known as Haemoproteidae. But, with the newer knowledge and tools, both the genera are now split into two or more genera or subgenera; *Haemoproteus* into *Haemoproteus* and *Paramoproteus* while *Plasmodium* into five subgenera, viz., *Plasmodium*, *Haemamoeba*, *Novyella*, *Huffia*, *Giovannolaia* and *Garnhamella*. Ideally, in case of *Plasmodium*, blood smears may contain two stages viz., gametocyte (of variable shape) and schizont (with merozoites of variable number) and only gametocyte, usually halteridium shaped, represents the blood stage of *Haemoproteus*. The problem lies if malarial blood smears represent only gametocyte stage (not schizont) and that too, of different shapes and sizes, puzzling species identification with the confusion of possible mixed infection

It is a common practice in parasitology that parasites are considered host group specific, if not species specific. Usually avian host family level specificity is widely accepted for haemoproteid parasites, extending also to the host order level, while in case of *Plasmodium* it is variable, usually occurring a wide host range, sometimes restricting to passeriform or galliform birds. In columborid birds the most common haemoproteid parasite is *Haemoproteus columbae*, the gametocytes of which usually extend along one side the host erythrocyte

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curving its ends, while the gametocytes of *Haemoproteus sacharovi* completely fill the host cell. The plasmodid species *Plasmodium relictum* is basically rounded parasite in erythrocyte, naturally occurring in different host orders comprising of 51 families and 270 species. Contrastingly, *Plasmodium elongatum* is elongated parasite, naturally occurring in a few host orders, comprising of 17 families and 50 species including columborid hosts. These two malarial species originally described from the type hosts *Passer hispaniolensis* in 1890 and from *Passer domesticus* in 1930 respectively

Identification Tools and Techniques

Basic identification tools and techniques based on blood stages are standard key to the species and thin smear technique. Camera lucida drawings facilitating length, breadth and area measurements of the gametocytes and parasite nucleus, hypertrophy or atrophy of host cell and host cell nucleus as well as counting of pigment are essential for quantitative assessment on graph paper. Shape of the gametocyte essentially helps differentiation of the parasites and segregating them into groups. However, newer techniques and tools like ultrastructure, SEM study, cyto-chemical study, DNA barcode study may be approached in critical cases. For routine identification purpose malarial species should be identified based on contrasting and easily recognizable taxonomic criteria, preferably using modern taxonomic approaches/methods. Cross transmission study may be employed in the laboratory in complex and doubtful cases as far as possible. Comparing with named collection by experts would be of much help in this respect. So, a reference collection of named species may be developed for quick comparative study. However, numerical taxonomic and molecular taxonomic approaches may also be employed as and when needed.

Expert Comments

In response to my query on how to separate *Haemoproteus* from *Plasmodium* in the absence of schizonts during Ph. D. dissertation work, Prof. Gordon F. Bennett, Department of Biology & IRCAH, Memorial University of Newfoundland, Canada, on May 8, 1975, wrote it is hard to answer. However, he elaborated and emphasized as follows:

“To a greater extent, it depends on experience and a ‘feel’ for the parasites. In less subjective terms, I have normally found that haemoproteids have a denser staining cytoplasm, the cytoplasm is ‘coarser’, the parasite nucleus is more discrete and the pigment granules are blacker and larger than those in *Plasmodium*. I regret that I cannot be more precise than this, but there are some species of *Plasmodium* and *Haemoproteus* that are virtually indistinguishable on the basis of gametocyte alone.”

Similarly, in reply to my letter of April 3, 1975, Prof Norman D. Levine, College of Veterinary Medicine, University of Illinois, Urbana, USA, wrote that your work on avian blood parasites sound worthwhile and gave me his opinion on identification of blood parasites as follows:

“I think that complete identification (giving the correct species name) is important. It is not enough nowadays to say merely that you found *Plasmodium* sp., *Haemoproteus* sp., etc. The species name should be determined and given. This will require cross-transmission studies, I know, and they are sometimes hard to carry out, but they are badly needed. Many names have been given simply because the parasite was found in a new host. Such names may be correct, but they are more likely to be synonyms of other names that have been previously given to same parasite in some other host. We are learning more clearly that many blood parasites can be transmitted from one host to another, even within different families. It is important, therefore, not to give new names to already-named parasites.

A letter relating to identification of avian malaria parasite identification received from Prof Marshal Laird, Research Professor (Parasitology) & Director - Research Unit on Vector Pathology, Memorial University of Newfoundland, Canada, stated on this issue and about their microfiche as follows:

“The two species of *Plasmodium* connection with which you cite Bhatia's 1938 book have not gained general acceptance. I am enclosing a recent microfiche representing a consensus on the part of several of us here over which of the true avian malaria parasites should

be regarded as 'good' species. I am not at all sure of the explanation of your remark concerning 2-6 merozoites in a *P. circumflexum*-type infection. This sounds rather like double infection, probably involving one of the *P. vaughani* group."

Comments on identification problem of avian malaria parasite also received from Prof. Reginald D. Manwell, Professor (Emeritus) of zoology and Research associate, Syracuse University, New York are given hereunder:

"I am interested in the parasite you describe. However, I have discovered that plasmodia of the subgenus *Novyella* are often very difficult to identify. You should, if possible, inoculate canaries and-if they prove susceptible--- study the behavior of the parasite in them. The problem is discussed in my 1969 paper entitled 'The Problem of Species in *Novyella*' which I think was one I sent you. Your description suggests *P. vaughani* or *P. hexamerium*; possibly *P. tenue*".

Lastly, for better understanding of the problem of species identification including misinterpretation, the letter dated September 14, 1976 of Prof P. C. C. Garnham, a world authority on Haemosporidia, is quoted below:

"Regarding *Proteosoma moruony*, I should not like to express an opinion on your film without seeing it. If you pack it up very carefully and send it to me, I could examine and return it. It is certainly possible for the nucleus of haemoproteid to appear split up and be misinterpreted as a developing schizont. Reference your second paragraph, *Plasmodium gallinulae* is referred to my book, page 935, but unfortunately the name was left out of the index. Both this species and *P. herodiadis* should be removed to the genus *Haemoproteus*."

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