CHAPTER 2

Microneme Proteins in Apicomplexans

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Introduction

The invasive stages (zoites) of most apicomplexan parasites are polarised cells that use their actinomyosin-powered gliding motility or "glideosome" system to move over surfaces, migrate through biological barriers and invade and leave host cells. Central to these processes is the timely engagement and disengagement of specific receptors upon the regulated release of apical invasion proteins from parasite secretory organelles (micronemes, rhoptries). In this short review, we summarise recent progress on identification and functional characterisation of apical invasion proteins mobilised to the parasite surface from the microneme organelles. We have restricted our focus to Toxoplasma, Eimeria, Cryptosporidium and the nonerythrocytic stages of Plasmodium because these organisms have been the most intensively studied apicomplexans that invade nucleated cells and because invasion by erythrocytic stages of Plasmodium is covered in the next chapter.

Micronemes are the smallest of the apicomplexan secretory organelles that cluster at the apical end of the zoite. The number of micronemes varies enormously between different genera, species and developmental stages with those zoites displaying vigorous and extensive gliding or migration activity generally having the most. Thus, Theileria zoites, which are nonmotile, do not migrate and do not display active host cell invasion, have no micronemes;1 merozoites of Plasmodium, which neither glide nor migrate but rapidly and actively invade erythrocytes, have few;2 sporozoites and merozoites of Eimeria, which glide, migrate through intestinal contents and actively invade enterocytes have many;3 and Plasmodium ookinetes, which glide and migrate through the midgut epithelium of the mosquito, but do not classically invade host cells, also have many (and by contrast, do not have rhoptries).4 This long-standing correlation between micronemes and parasite motility, migration and invasion is well supported by a variety of biochemical and genetic studies which show: (1) that microneme secretion is rapidly up-regulated when parasites make contact with host cells;5 (2) that some Plasmodium microneme proteins are targets of erythrocytic invasion-inhibitory antibodies;6−10 (3) that parasite invasion is blocked when microneme secretion is chemically inhibited;11,12 and (4) that genes encoding MICs either alone, or in concert with others, are essential for effective parasite motility, migration and invasion.

MICs have been identified in a variety of approaches (reviewed in ref. 13-14), most recently through the application of proteomics to gradient-purified organelles and excreted-secreted antigens.15,67 Figure 1 summarises the current repertoires of MICs, including only those genes for which a full sequence and a verified organellar localisation is known. The majority of MICs comprise multiple copies of a limited number of adhesive domain types, which has allowed the identification of a large number of additional putative microneme
Figure 1. Modular MICs. Schematic representations of known microneme proteins from four different apicomplexan genera are depicted (not to scale). Accession numbers, where available: *Eimeria tenella* MIC1, M73495; EtMIC2, Z71755; EtMIC3, AAR87667; EtMIC4, CAC34726; EtMIC5, AJ245536; *Cryptosporidium parvum* TRAP-C1, AAB92609; GP900, AAC98153; CpSCRPA061328; *Plasmodium falciparum* TRAP, AAC1867; *Plasmodium berghei* SPECT, BAD08209; PbsSPECT2, BAD83404; PbCeTOS, BAD97683; PbSOAP, AAL07530; PbCHT1, CAC40151; PbsWARP, AAK83296; PbMOAP, AAV28504; PbCTRP, AAF73158; *Toxoplasma gondii* MIC1, CAA96466; TgMIC2, AAB63303; TgM2AP, AAK74070; TgMIC3, CBB54444; TgMIC4AAD3906; TgMIC5, CAA70921; TgMIC6, AAD28185; TgMIC7, AAK35070; TgMIC8, AAK19757; TgMIC9, AAK19758; TgMIC10, AAG32024; TgMIC11, AAN16379; TgMIC12, AAK58479; TgAMA1, AF010264; TgSUB1, AAK94670. A color version of this figure is available online at www.eurekah.com.

proteins bearing these domains in the parasite databases. Based on this it is likely that many more proteins will be shown to occupy the micronemes in future studies.

**Ligand Domains and Their Cellular Receptors**

**Thrombospondin-1 Type 1 Domains (TSR)**

Thrombospondin-1 (TSP-1) is a multifunctional, glycoprotein adhesion molecule that mediates a broad range of biological interactions via three distinct repeated domains designated types 1, 2 and 3. The adhesive TSP-1 type 1 domain, TSR, is a small ~60 residue structure found in the extracellular regions of several protein families involved in immunity, cell adhesion and neuronal development, and shown to have binding activity for a number of cellular and matrix molecules (reviewed in ref. 19). The TSR is an ancient eukaryotic module that is found in many nematode and arthropod proteins as well as those from the