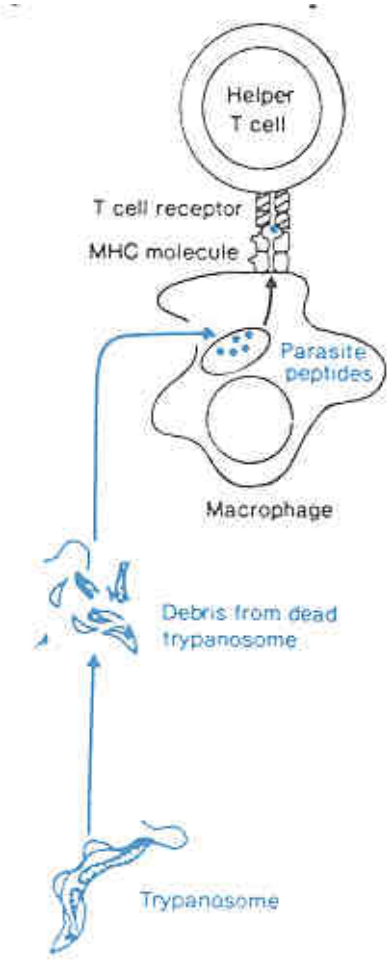

This simplified diagram illustrates the steps by which bovine T lymphocyte cells recognize antigen.

Peptide molecules of degraded antigens are presented to T lymphocytes on the surface of antigen-presenting cells, such as a macrophage, in a binding cleft of a bovine MHC molecule. T cells recognize antigen only when it is associated in this way with a 'self' MHC molecule.

Types of MHC molecules vary among individual animals. ILRAD scientists need methods with which to characterize these MHC molecules to determine if this polymorphism will affect the efficacy of the novel p67-based vaccine against East Coast fever now under development at the Laboratory.

Ph.D. Theses

A *Trypanosoma congolense*-specific antigen released in the course of an infection is identified as a thiol protease precursor

Standardized reagents, including trypanosome antigens, are needed to support research in the epidemiology of trypanosomiasis. A peptide doublet of *Trypanosoma congolense* with a molecular weight of 38/40 kiloDaltons (kDa) and recognized by diagnostic monoclonal antibodies was used to raise polyclonal antibodies. One of these antibodies was used to screen a complementary DNA (cDNA) library from which 14 positive clones were identified. Antibody subpopulations selected by 10 of the 14 clones specifically recognized the 30/40-kDa polypeptides in lysates of different isolates of *T. congolense.* These recombinant antigen-selected antibody subpopulations appeared to have retained the species-specificity of the original polyclonal.

A clone with a cDNA insert of 1.6 kilobases, which had hybridized with all the other positive clones, was further characterized and sequenced. Results of a time-course digestion of *T.*

congolense DNA with restriction enzymes revealed that the gene encoding the *T. congolense* species-specific antigen occurs as an imperfect tandem repeat in the genomes of these parasites, with an estimated copy number of between 20 and 30.

The gene appears to be polymorphic among three isolates of *T. congolense.* A complete nucleotide sequence of the cDNA was obtained. Analysis of the whole of the deduced amino acid sequence revealed a number of domains identified as potential signal sequences. A homology search in databases revealed a high degree of identity of the deduced protein sequence to cysteine proteases from both animal and plant sources.

The gene is one of two studied that encode antigens secreted by trypanosomes or released by the parasites on dying into the blood and other tissues of mammalian hosts. Characterizing these antigens and making them available will offer scientists standardized reagents for use in studies of trypanosome epidemiology.

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Summary of a thesis abstract presented for a Ph.D. granted in 1993 by the Department of Biology and Biochemistry, Brunel University, UK.
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An extra chromosomal DNA molecule of *Theileria parva* encodes mitochondrial protein

Theileria parva, a protozoan parasite of cattle, contains an extra chromosomal DNA molecule with an apparent mobility of 7.1 kilobases in agarose gels. This molecule was characterized to determine whether it played a role in parasite virulence or, more importantly, in the unique ability of *T. parva* to 'transform' bovine lymphocytes. In addition, as an extra chromosomal element, it was potentially a good starting point for the construction of transfection vehicles.

A total of 5893 base pairs of this extra-chromosomal molecule have been fully sequenced and analysed. The molecule is linear in structure and contains inverted repeat sequences at its telomeres. The sequence shows that the DNA may code for apocytochrome b and polypeptide I and III of cytochrome c oxidase, protein components of the last two mitochondrial electron transport complexes. Apocytochrome b is of interest because it may be the target of hydroxynapthaquinones the class of drugs used in chemotherapy for East Coast fever.

The DNA molecule also contains several unique short stretches of discontinuous sequences that are similar to both large (LSU) and small (SSU) ribosomal subunit RNA of *Escherichia coli.* Five fragmented ribosomal RNA-like sequences, LSU1-LSU5, can be folded by inter-and intra-molecular base pairing into the phylogenetically conserved domains IV and V of the LSU ribosomal RNA. Other short stretches of sequence with similarity to SSU rRNA and other regions of LSU rRNA were tentatively identified. ,

The protein coding potential of the DNA molecule is characteristic of mitochondrial DNA and its structure and the presence of scrambled rDNA sequences is reminiscent of the mitochondrial DNA of *Chlamydomonas reinhardtii.* Other protozoan parasites in the same phylum as *Theileria,* such as *Plasmodium, Babesia, Eimeria* and *Toxoplasma,* contain related DNA molecules. These molecules are different in structure to the *T. parva* molecule but the conservation of the coding potential argues for a functional significance of this family of related DNA molecules.

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