PROSPECTS FOR VACCINES AGAINST NEMATODE PARASITES OF SHEEP

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Introduction

There are no commercially available vaccines for the control of sheep nematodes. They are nearly always controlled by a combination of anthelmintic drugs and pasture management. Although this practice can be extremely effective, it is probably not sustainable, as the situation is threatened by anthelmintic resistance. In the last decade considerable progress has been made in identifying candidate vaccine antigens for several of the important species. This review concentrates on some of the strategies that are being used to achieve this goal and provides a list of the various protective antigens that have been discovered.

Vaccine strategies

In the 1960s it was discovered that infection with *Dictyocaulus viviparus* larvae which had been attenuated by irradiation could stimulate a high degree of protection in calves against natural challenge. This led to the lungworm vaccine that is still sold today. Attempts were immediately made to extend the principle to the ovine gastrointestinal nematodes, but unfortunately it was found that while the method worked well for *H. contortus and T. colubriformis* in sexually mature sheep, the protective effect was too weak in lambs. Of course, no vaccine can be commercially successful if it is not efficacious in young, highly susceptible lambs on pasture.

Research is being conducted into the mechanisms of naturally acquired immunity to gastrointestinal helminth infections of sheep, but it is still not known what the final effector mechanisms are. The situation seems to be complex, involving a combination of local hypersensitivity, cell mediated, antibody and inflammatory responses and is complicated further by the natural unresponsiveness that exists in the young lamb and in the ewe around parturition. Only when these mechanisms and the respective antigens that trigger them are finally unravelled, can rational attempts be made to artificially stimulate them.

The vaccine strategy currently enjoying most success largely ignores the mechanisms of natural immunity, but attempts to direct responses towards potentially susceptible targets on or secreted by the parasite. Antigens that are recognised by the host during the course of infection are termed “natural” or “conventional”. However, in the case of blood feeding *Haemonchus* the luminal surface of the intestine has been a particularly rich source of suitable target molecules. Because they are not recognised immunologically by the host during infection, these antigens are classified as “hidden”, a feature that raises some unconventional concepts relating to vaccination strategy.

The general approach for either type of antigen has been:

1. To screen candidate protective fractions enriched for the parasite target in preliminary protection trials,
2. To purify the protective components as much as possible and
3. Finally to isolate and express the genes which encode these so that a functional recombinant protein can be produced.

Of course, the final step of producing a functional recombinant antigen is crucial to the whole approach, because a vaccine that consisted of native worm antigen could never be a commercially viable proposition.

Natural antigens

Striking protective effects have recently been reported against *Haemonchus,* using two proteins from adult worm excretory-secretory products that are recognised serologically by infected sheep (Table 1). The nature and biological function of these molecules has yet to be determined, but it will be interesting to see whether these antigens are effective in the young lamb and periparturient ewe where natural immunity is poor.

Hidden antigens

The mechanism is straightforward in the case of blood feeding *Haemonchus.* The sheep is immunised with nematode gut membrane proteins and a high titre circulating antibody response is raised. When the worm subsequently feeds, it ingests antibodies which bind to functional proteins on the brush border of its intestinal cells, so that its digestive processes are compromised, leading to starvation, loss of fecundity and weakness. Eventually the parasite becomes detached and is swept out of the gut by peristalsis. Several different protective gut membrane proteins have now been isolated (Table 1). Most of these have been identified as various proteases that are presumably involved in the digestion of the blood meal.

A major advantage of the hidden antigen approach is that, because the mechanism of immunity is quite different from normal, it works in situations where natural immunity to *Haemonchus is* weak or ineffective. Thus it has been shown that young lambs and periparturient ewes can be successfully immunised. On the other hand, the fact that the antigens are hidden means that, unlike conventional vaccines, immunity is not boosted by infection. At first sight it might seem that frequent immunisations would be required for an effective level of protection to be maintained. However, the mechanism of protection in lambs immunised by this method mainly affects late fourth stage and older worms, whereas incoming larval stages, which are not yet blood feeders, are largely unaffected. Thus, when vaccinated lambs were given repeated daily infections of larvae, to mimic the situation in the field, faecal egg counts were controlled by the vaccine but the continued presence and activity of the early larval stages stimulated a natural immunity which replaced the vaccine immunity. It is hoped that transmission of Haemonchosis will be controlled both by vaccinating lambs before weaning and ewes during pregnancy.

Another potential advantage of hidden over natural antigens is that, because the host does not respond to them during natural infection, the parasite has not needed to adapt to counteract the host response. Thus hidden antigens are likely to be conserved, both within and probably between genera.

It is not entirely clear whether the gut antigen principle can be successfully employed against species of

Table 1.
Candidate gastro-intestinal nematode vaccine antigens which have given substantial protection.

Parasite	Antigen (hidden/ natural)	Type	Mol. Wt. (kDa)	Gene(s) isolated	Identification / function	% Protection eggs
H. contortus	H11	H	110	3	aminopeptidases	94
	H-gal-GP complex	H	91, 41 42, 40	4	Metallopeptidases pepsin / pepsinogen	93
	Thiol binding	H	40 - 55	1	Cysteine proteases	92
	GA1	H	46, 52	3		50
	H45 or P150 complex	H	45, 49, 53	1		96
	ES15+24	N	15 and 24			73
O. circumcincta	ConA	H	various			80
	Thiol	H	?			71

nematode which are not direct blood feeders. The precise diet of economically important species *like Ostertagia* is not known, but they do contain host immunoglobulin, presumably ingested with mucus, tissue fluids and / or serous exudates. It remains to be determined whether the amounts consumed are adequate for this vaccination strategy to be effective, but there are at least two encouraging results with *Ostertagia circumcincta.*

Vaccine formulation and mode of delivery

The choice of adjuvant or delivery system can be vital for the success of any vaccine. In the case of the nematode hidden antigens, an adjuvant that stimulates a prolonged high titre antibody response is clearly what is needed. For those species of nematode which reside in the small intestine, it may be necessary for a recombinant vaccine to stimulate facets of the mucosal response for effective protection to be induced. The possibility of using *Salmonella* vectors for achieving this is actively being pursued. The concept of naked DNA vaccines is attractive particularly because it could short circuit the need for producing functional recombinant proteins, but preliminary experiments with sheep have not been encouraging.

How good do worm vaccines have to be?

Conventional wisdom suggests that a worm vaccine would have to approach the efficacy of either anthelmintics or bacterial and viral vaccines to be effective. In the case of the gastrointestinal nematodes of sheep it is more appropriate to consider a vaccine as an epidemiological tool to maintain low level pasture contamination, rather than a weapon to abolish infection completely. Respected mathematical modellers have concluded that a vaccine containing natural antigens need only have 60% efficacy in 80% of the flock to bring substantial benefits. Further, if the vaccine was based on hidden antigens, then protection of 80% of the flock with 80% efficacy would give better control than a conventional anthelmintic programme.

Conclusions

Considerable advances towards nematode vaccines for sheep have been made in the last decade. Several highly protective antigens have been discovered and the genes encoding several of these have been cloned. However there are still considerable hurdles to be overcome before a monovalent vaccine *for Haemonchus* reaches the market place and the prospects for a multivalent nematode vaccine are more distant. The main obstacle is the production of cost-effective recombinant versions of the protective antigens. However, if the rate of progress that has been achieved over the last ten years can be sustained, then the next decade should see the launch of the first defined antigen nematode vaccine for sheep.

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