Experiences with a safe live Pasteurella multocida *vaccine*

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Introduction

Recent advances in understanding of the LPS structure of PM isolates has explained the limitations of Heddleston serotyping in predicting protection by killed vaccines (Harper and Boyce 2017). There are currently 8 recognised LPS genotypes with the main chicken genotypes being in either L1 (Heddleston serovars 1 and 14) or L3 (Heddleston serovars 3, 4 and 3/4) or L6 (Heddleston serovars 10, 11,12, and 15). Obviously just getting the result L1 from a farm investigation can tell you that if you are using a L3 killed vaccine that it will probably not work (but gives no guarantee that a L1 killed vaccine will work). Even within a Heddleston serovar some variations in LPS structure will be not generate homologous protection (see Morrow 2017). This explains why autogenous vaccines are initially successful, but the field strain may change by becoming an escape mutant. There is some evidence emerging that this ability to change LPS structure (within a LPS genotype) may be an innate property of PM strains used for immune evasion during natural challenge. Antibody responses to killed vaccines are generated to LPS antigens remaining immunogenic after inactivation. Late breaks in lay in autogenous vaccinated flocks may be due to the duration of immunity being insufficient.

Live PM vaccines generate broad immunity within and across LPS types and probably includes CMI to protect against fowl cholera (FC). The current problems with traditional US live vaccines are reversion to virulence or residual virulence (Blackall and Hofacre 2020). In Australia we have been using an *aroA* mutant PM vaccine by injection (Parent strain X73 US Heddleston serovar 1 type strain, Homchampa et al 1997). This safety attenuation is particularly effective in chickens (but not turkeys) as this strain (PMP-1) only multiplies in the laboratory and not in the bird or the environment after exhaustion of aromatic amines and cannot be found in the chicken after 2 weeks. Duration of immunity in the lab to homologous challenge and heterologous challenge has been limited but, in the field, it seems to be adequate (the laboratory challenge models that demonstrated efficacy used very strong challenges. What we need now are reports of efficiency in the field). This paper aims to report current efficiency results.

Materials and Methods

Users who have used Vaxsafe PM as registered or in live/killed formats were asked for comments.

Results

65000 free range layer farm. (1000 bird flocks in movable arks).

Flocks are vaccinated with Vaxsafe PM by intramuscular injection at 4 and 8 weeks of age during the vaccination for Eryvac, MS and MG (4 weeks) and again with Eryvac, EDS/ND, fowl pox, AEV and ILT SA2 at 8 weeks. The vaccine is reconstituted in Mareks Diluent and one dose is contained in 0.2 ml injection into the breast muscle. Some diluent is initially drawn into a syringe

and then injected into the vial where the freezed dried vaccine plug is dissolved. It is then removed from the vial and injected into the diluent bag.

The owner commented *"Every new flock that had been vaccinated did not develop the issues we were having in the older flocks vaccinated with other vaccines. Once the last of the older flocks had been culled out we had no issues and no need for medication. Then we had the MG and MS issues. The MS MG vaccine stopped that issue, Recently however we have seen the MG MS issue arising again although with milder mortality rates that don't seem to be escalating like they did before. We have started treating some flocks with CTC."* The young vaccination age is because the contract vaccination team is not local. The flocks are reared in a traditional fixed shed.

Broiler breeder flocks on a "chicken sick" site.
An integrator with a production farm that traditionally had a PM problem that had lately been controlled by killed vaccines reared a flock and had no vaccine available. Even though FC had not been seen for 10 years in this operation they had an outbreak in the unvaccinated flock. The size of the flock was 10K females and the amount of PM vaccine needed was too small for economic autogenous batches. The next flocks were vaccinated twice with Vaxsafe PM by intramuscular injection and no problems have been seen in the next 10 flocks. No antibiotics have been used on these flocks for FC since.

To the best of my knowledge no other producers have used the vaccine as it is registered. Most use in the field is as a primer for killed autogenous vaccines. There is no published experimental studies or field studies on the effectiveness of this approach (in the USA killed vaccines are often followed by live vaccine injection on the theory that this may protect against pathogenic effects from their live vaccines). There has been some investigation by users if the live PM vaccine can be mixed in with killed vaccine (EDS/NDV) but the consensus has been to give it separately. Some people have used double barrelled syringes or twin injection machines to minimise handling of the birds.

The Vaxsafe PM vaccine has poor efficacy in laboratory studies when given orally (30% protection) (Scott et al 1999) and in the field its sensitivity to chlorine in drinking water would also need to be protected against. Some farmers have given it in water in the face of an outbreak in flocks vaccinated with live/dead or autogenous PM vaccines (trying to maintain organic status) have seen benefits while others have seen no benefit. Chlorine as the explanation of this variation is being investigated currently.

Conclusions

FC in layers has re-emerged as an important disease as we have taken birds out of cages. Outbreaks of FC in a 40K flock of commercial cage free layers at 40 weeks of age in Australia have recently been estimated to be $0.5M in lost production and disruption; let alone mortality, cost of diagnosis, cost of vaccination and cost of medication. Current vaccination with single live and/or single or multiple dead vaccinations have not been total solutions. There has been a reluctance to use Vaxsafe PM as registered (two injections 4 weeks apart) due to the published efficacy and label claim (http://bioproperties.com.au/!Pages/Vaccines/VaxsafePM.html) but the efficiency in the field seems good and warrants further trialing.

Serology using Guildhay PM kits is recognised as poor for monitoring vaccine response to either live or the killed vaccines (but large responses are seen to field outbreaks) (Peter Scott *pers. comm* 2022). The duration of immunity of killed vaccines may be the explanation for the outbreaks in the field in killed vaccine older flocks. Traditionally outbreaks of FC in Australia have been in

autumn in sheds with dirt floors– some conjecture on the mice coming in from the harvested fields as the days got colder. Standing water on the ranges appears to be a risk factor for free range flocks. The birds after vaccination are often depressed for one day and this could slow growth in the weeks that the birds are vaccinated. I consider this a measure of vaccine take.

The implications for the issuance of future Permits by APVMA for PM autogenous vaccines are obvious. Users will have to use the Vaxsafe PM vaccine by the registered method to make a case for issuing the permit after demonstrating that the vaccine has failed on their farm (presumably reported to the APVMA). Just showing that the challenging field strain is a L genotype similar to that in a permitted autogenous vaccine will not be enough (as such a result is not a predictor of protection (Harper and Boyce 2017)).

This is a major advance in protection from fowl cholera for chickens and maybe other species. Overseas live PM vaccines have enormous problems with reversion to/residual virulence (Blackall and Hofacre 2020) and would not be registerable in Australia. The crippling of X73 by the *aro*A deletion means that the vaccine has not been found after 14 days in birds (or the environment). We need another vaccine for turkeys. We also need people to gain more field experience starting from the registered use.

References

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