VACCINE RESEARCH INTERNATIONAL Plc

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Vaccine against Staphylococcal infections

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Staphylococcal infections

- Staphylococcus aureus is responsible for a variety of infections
- Post operative (wound, failed joint replacements)
- Infections in dialysis patients
- Osteomyelitis

Resistance

Normally treated with antibiotics

Resistance to antibiotics is a problem

Methicillin resistant Staphylococcus aureus (MRSA) is a big problem in UK, USA, Europe and Japan

More recently reports of Staphylococcus aureus with reduced sensitivity to vancomycin, the drug of choice for MRSA has introduced a note of urgency to deal with these infections.

Importance

- Hospital acquired infections
- 9% incidence of patients are infected at any one time
- 18.5% due to Staphylococci
- Over 40% of these are MRSA

Vaccination

Antibiotics have the problem of resistance

Vaccination to prevent infection will be a critical strategy

Resistance to antibiotics may be irrelevant to successful vaccination

Staphylococcal vaccines 1

Group and Year	Type of vaccine	Immunisation schedule	Infection	Results
McCoy and Kennedy 1960	Autologous	Intracutaneous 2-3 days apart total of 10 injections	Recurrent skin infections, osteomyelitis, post-operative wound infections, septicaemia	44/60 Excellent improvement 11/60 Improvement 5/60 Failure
Dillenberg and Waldron 1963	Polyvalent somatic antigen	Intramuscular 2 doses at 6 week intervals	Impetigo	Reduction in boils and impetigo in 90% vaccinated subjects in placebo controlled trial

Staphylococcal vaccines 2

Group and Year	Type of vaccine	Immunosation schedule	Infection	Results
Salmon and Symonds 1963	Staphylococcal bacteriophage lysate	Intranasal alternate day for 10 days then weekly and monthly	Chronic Staphylococcal infection, eczema, upper respiratory, pyoderma	486/607 Recovered 110/607 Improved 11/607 Not helped
Bryant et al 1965	Staphylococcal bacteriophage lysate	Intramuscular injections	Recurrent furunculosis	No significant difference between vaccinated and control groups

Staphylococcal vaccines 3

Group and year	Type of vaccine	Immunisation schedule	Infection	Results
Mudd et al 1965	PVL toxoid Divasta- Institut Pasteur – Alpha toxoid Staphage – Delmont Laboratories	Patients received Leucocidin +Staphage or Staphage+ alpha toxoid	Osteomyelitis and wound infection	Vaccinated patients did better than non vaccinated. Optimal leucocidin response in 3-5 weeks. Increase in anti-alpha toxoid antibody. No controls
Poole Warren et al 1991	Staphypan Berna – Mixture of inactivated Staphylococci and toxins	6 intramuscular injections of increasing concentration	Prevention of infection in CAPD patients	124 patients; no difference in incidence of peritonitis or catheter related infections between vaccinated and control subjects

Staphylococcal vaccines 4

Group and year	Type of vaccine	Immunisation schedule	Infection	Results
Shinefield et al 2002	Capsular polysaccharide 5 and 8 conjugated vaccine	Single intramuscular injection	End stage renal dialysis patients	Reduction in bacteraemia for up to 40 weeks post vaccination
Ahmad et al 2006	Chloroform inactivated whole Staphylococcus aureus vaccine	4 subcutaneous vaccinations at 2 week intervals	Phase I trial placebo controlled double blind	All vaccinated patients demonstrated increase in immune response
Merck	Staphylococcus aureus iron surface determinant B surface protein	Intramuscular with aluminium hydroxy-phosphate sulfate adjuvant	Phase I and Phase II clinical trials	Immune response in humans and macaques and protection in murine model

SA 75 Staphylococcus aureus vaccine

- **Method of vaccine preparation**
- The Vaccine is a chloroform-inactivated preparation of Staphylococcus aureus P/DFO 75
- Whole cell vaccine may offer cross reactivity with other strains, species and genera of bacteria.

Toxicology

Animals were observed for local and systemic effects post vaccination

Single dose Toxicology in 10 rabbits (subcutaneous – High Dose)

Repeat Dose Toxicology in 36 rabbits (Groups of Rabbits given 4 doses at 2 week intervals – High and Medium Dose)

Intramuscular and Subcutaneous in 12 rabbits (High Dose)

AMES test for bacterial mutagenicity

Phase I Clinical Trial

Trial conducted by Simbec, Merthyr Tydfil, Wales

Placebo controlled double blind

48 male volunteers

3 Dose Groups with 12 test and 4 placebos in each
Dose escalation, evaluation of safety data after two vaccinations for each group.

0.15mg per dose
0.36mg per dose
0.45 mg per dose

4 vaccinations given subcutaneously in 0.5ml volume at 2 week intervals

Vaccine manufactured under GMP by Norwegian Institute of Public Health

Clinical Trial

Safety and Immune response

Volunteers observed for local and systemic signs (erythema,induration,bruising, pain, ECG, blood pressure, temperature, haematology, biochemistry) at 0hr, 4hrs and 8 hrs on the day of vaccination (day 1) and days 2, 3 and 8

Blood samples taken before and after each vaccination for immune response evaluation.

Completion of study

One subject in 0.15 mg group did not complete the study

One subject in the 0.36 mg group left as he moved away

One subject in the 0.36 mg group withdrew himself due to flu like symptoms

One subject in the 0.45 mg group was withdrawn from the trial after one vaccination due to severe injection site erythema.

Safety Summary

Local Adverse Events

(Injection site Erythema, induration, harmorrhage,burning,pain, pruritis, swelling, warmth)

Placebo	6
0.15 mg Vaccine	117
0.36 mg Vaccine	145
0.45 mg Vaccine	197

Safety data (2)

- Non local Adverse Event

(fatigue, feeling hot or cold, flu like illness, malaise, vomiting, headaches)

Placebo	23
0.15 mg Vaccine	29
0.36 mg Vaccine	19
0.45 mg Vaccine	63

Immune response

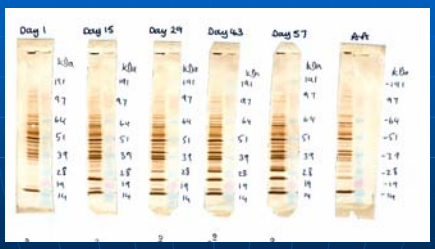
Immune response was evaluated by testing sera from volunteers before vaccination and after 1, 2, 3 and 4 vaccinations by Immunoblotting and ELISA against homologous vaccine preparation

ELISA against four binding proteins namely collagen binding protein, fibronectin binding protein, fibrinogen binding protein and extracellular adherence protein

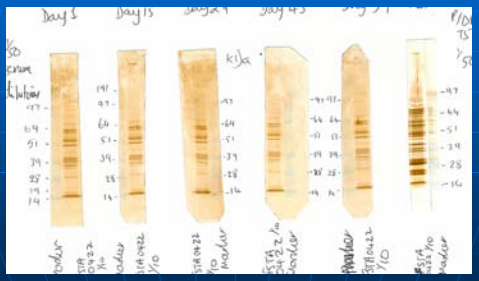
Summary of IgG ELISA results and antibodies to binding proteins

Subjects in Trial	ELISA Doubling (1/50) Number of subjects	Collagen binding protein Number of subjects	Fibrinogen binding protein Number of subjects	Fibronectin binding protein Number of subjects	Extracellular adherence protein Number of subjects
Placebo	1/12	1/12	2/12	0/12	0/12
0.15 mg Vaccine	8/12	6/12	0/12	0/12	1/12
0.36 mg Vaccine	6/12	9/12	0/12	0/12	1/12
0.45 mg Vaccine	9/12	7/12	2/12	0/12	0/12

Immunoblot of sera from subject (038) given 0.36mg Vaccine



Immunoblot of sera from subject (006) given placebo



Summary of IgG Immunoblotting data

Subjects	No reaction	Weak reaction	Strong reaction
Placebo	7/12	5/12	0/12
0.15 mg Vaccine	0/12	6/12	6/12
0.36 mg Vaccine	0/12	2/12	9/12
0.45 mg Vaccine	0/12	0/12	12/12

Conclusions

- 0.15 mg and 0.36 mg doses of vaccine are considered safe and well tolerated
- 0.45 mg represents the upper limit of the dose concentration
- The data shows significant immune response following the first vaccination at each dose level with increased reactivity after two, three and four vaccinations
- The data suggests it may be possible to reduce the number of vaccinations required to obtain an optimal immune response
- Further testing of sera for opsonic antibody towards possible correlation with markers of protective efficacy.

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