Full Consent Application for the Meningococcal GroupB OMV vaccine MeNZB®, by Chiron Vaccines at the Rosia and Siena Sites, Italy Application in Accordance with Medicines Act 1981 & Medicines Regulations 1984, for Consent to distribute in New Zealand

Reviewer:

July 10th 2006

Data is presented in 5 Modules, subdivided into 53 volumes.

Module 1: Contains:
The application for full registration:
Certificates of approval of Good Manufacturing Process for both Rosia and Siena sites, issued by the Italian Medicines Agency as established by European Union and recommended by World Health Organisation. Last authority inspection 7th – 10th April 2003.The Data Sheet, in a revised version from that approved in July 2005A Patient Information Sheet (CMI) dated August 2005
Labelling information including examples of labelling for individual vaccine files and the secondary labelling for the outer package.

Comment: The Data Sheet has been modified to accommodate delivering the primary immunization course of 3 doses at age 6 weeks, 3 months and 5 months to infants aged less than 6 months, as for other vaccines in the NZ schedule. There appears to be no other change from the version approved previously.
Some of the terminology used in the Patient Information Sheet may be rather too technical for lay persons – e.g. "antero-lateral thigh", "deltoid region" "booster dose", "neurological reaction", "excipients".
The labelling conforms to the NZ regulations

Module 2 Vol 1: This contains a series technical document summaries, revised in December 2005, giving details of vaccine manufacturing process, controls, reference standards, pharmaceutical development and controls, nature of the excipients and containers, the manufacturing facilities and safety testing.

Module 3, Vols. 1, 2, 3 & 4: This module gives more detailed information on the selection of the MeNZB antigen, the process of development of the vaccine in association with the Norwegian National Institute of Public Health, (NIPH) and comparisons of MeNZB with the Norwegian parent vaccine MenBvac.

Module 4, Vols 1, 2, 3, 4 & 5: These volumes contain reports on the immunogenicity of various concentrations of antigen given to mice and NZ white rabbits in single and multiple doses. Studies of toxicity have also been carried out in mice and rabbits with specific studies in rabbits on the effects of multiple doses of vaccine given both before and during pregnancy with examination of the foetuses for any ill effects.

Note: Summaries of the studies contained in Modules 2, 3 and 4 will be presented at the end of this report.

Module 5, Vols 1 – 18 contain details of the clinical studies carried out with both the Norwegian vaccine MenBVac® and the New Zealand vaccine MeNZB® (formerly called NZ MenB, but to avoid confusion this reviewer will us the name MeNZB throughout this report). Many of the clinical studies have been reviewed by the

MedSafeVaccines Subcommittee previously and will only be referred to briefly, although some have been subject to review. Other study reports are new and are summarised in greater detail.

Final reports of the **Independent Safety Monitoring Board** (dated May 23rd 2006) and the **Meningococcal B Vaccine (MeNZB®) Effectiveness Assessment** have been circulated separately.

Module 5 Vols 1 – 18:

Table Module 5,Vol 1 5.2, is a Summary of the Clinical Studies reported.

Vol. 1: This module also contains a report (Version 2, dated September 2003) on the development and validation of the SBA assay in an interactive series of studies between the Manchester PHLS, Chiron Laboratories at Emeryville, the Norwegian NIPH and ESR (NZ) using 4 target strains of N. meningococcus B, the NZ vaccine strain NZ 98/254, the Norwegian vaccine strain 44/76 SL and two NZ wild strains NZ94/167 and NZ02/09. This study showed considerable variation in the estimation of the absolute SBA titre for the same sample between the 4 laboratories but greater consistency when expressed as ≥ 4-fold rise in titre from baseline and consistency in results within the individual laboratory on repeat testing (≥ 95% at ESR). Much of the variation may be due to differences in the source of complement used in the test. SOP (Standard Operating Procedure) established at Chiron. Now use a blended serum complement and NZ98/254 and NZ02/09 as antigen in all clinical trials. Three clinical trials, V60P1, V60P2 and V60P3 showed differences in both homologous and heterologous SBA responses between individuals.

Vol. 2A, Study V60 P1 (Final Report dated 19th June 2003 – no update expected, but note new study V60 P1E1 reported below in Vol.8).
Phase 1 / 2 study in adults – previously reviewed. 75 adults aged 18 – 50 yrs in 3 groups given 25µg, 50µg MeNZB or 25µg MenBVac. 78-88% adults showed ≥4 fold SBA response to MeNZB vaccine after 3 doses at 6-week intervals when tested against NZ98/254 but only 44% when tested against MenBVac (44/76 SL). Adverse events, mainly pain and redness at injection site.

Vol. 3A Study V60P2: (Final Report dated 25th March 2004 – no update expected). Randomised, blind study to evaluate safety, reactogenicity and immune response to 3-dose schedule in 8 – 12 yr old children. Three groups given either, NIPH produced MeNZB, Chiron produced MeNZB or MenBVac. 79% of those receiving Chiron produced MeNZB showed ≥ 4-fold increase in SBA titre compared with 73% after three doses of NIPH MeNZB and 32% after receiving MenBVac. SBA GMT was 24 in the Chiron vaccine group, 4 weeks after the 3rd dose and 22 in the NIPH vaccine group. Adverse reactions were similar in 20 – 40% of all groups with local reactions predominating. There was no significant difference in the immune response to either the Chiron or NIPH produced MeNZB.

Vol. 4 A: Study V60 P3: (Final Report dated 8th January 2004 – no update expected) Phase 2, single centre; blind randomised study to evaluate safety, reactogenicity and immunogenicity of either MeNZB or MenBVac to 16 –24 month old toddlers. After 3rd dose 75% of those receiving MeNZB showed ≥ 4 fold rise in SBA titre compared with 4% of those receiving MenBVac. The % showing a titre of ≥1:4 was 92% for the MeNZB recipients. SBA GMTs 4 weeks after the 3rd dose were 16 and 1.56 for the

MeNZB and MenBvac recipients respectively. Local reactions with redness and swelling in 80 –90% and systemic symptoms of irritability, sleepiness and change in appetite in 30-40% were noted in those receiving MeNZB.
Comment: Unlike the older age groups these toddlers showed little cross reactivity in response to the two vaccine antigens, NZ 98/254 and Norwegian 44/76. This suggests that they would have less protection against Meningococcus strains other than the predominant epidemic strain in NZ.

Vol. 5A: Study V60P5: (Final Report dated 11th October 2004 – no update expected).
A phase 2, single centre, blinded, randomised study to evaluate the safety, reactogenicity and immune response to MeNZB when administered to infants aged 6 – 8 months. Two groups of infants, the first given MeNZB, the other given Menjugate® Meningococcal C conjugate. 74% of those receiving MeNZB showed ≥ 4 fold increase in SBA titre 4 weeks after the 3rd dose of MeNZB and 92% had titre of ≥ 1:4. ELISA GMC increased 64 fold. Those receiving the Menjugate® showed minimal response to NZ98/254 antigen. Adverse events occurred in up to 60% but were not severe.

Vol. 6A: Study V60P6: (Report dated 15th December 2005 – final report).
A phase 2, single centre, randomised, observer blind, controlled study to evaluate the safety, reactogenicity and immunogenicity of Chiron MeNZB vaccine when administered concomitantly with routine immunization vaccines to 6 – 10 week old healthy infants.
The objectives of the study were to evaluate the immune response to the Chiron MeNZB 6 weeks after a 3rd dose and the response 6 weeks after a 4th dose given at age 10 months in a subset of infants.
A secondary study was to evaluate, 6 weeks after the 3rd dose of MeNZB, the immune response to the routine vaccines Hib, DTaP, IPV and Hep B used in the NZ schedule, either given concomitantly with MeNZB or separately.
Methods:
375 infants were enrolled and divided into 2 groups of 250 and 125 respectively. They the first group of 250 received MeNZB + routine vaccines at 6 weeks, 3 months and 5 months and a subset of 51 infants from this group received a 4th dose at 10 – 11 months. The second group of 125 infants received routine vaccines. Both sets of infants received an equal number of injections at each immunising session, to maintain blinding. This required some variation of the routine vaccines used in each group, thus Group 1 received DTaP-IPV (Infanrix-IPV® - GSK), Hib-HBV & (Comvax®- Merck) +MeNZB while the second group received HBV (HepBvax®- Merck) DTaP (Infanrix® - GSK) and IPV (IPol® - CSL-Aventis). So that local reactions could be monitored, the MeNZB was always given into the right thigh in Group 1 and the other vaccines into the left thigh. In Group 2 the DTaP vaccine was always given into the right thigh, the other vaccines into the left.

See: **Subject Completion Flow Chart, Attachment 2 CSR V60 P6 (15 June 2006)**

The chart shows that, as would be expected, a number of the infants did not complete the study – final numbers are 243 in Group 1, of whom 46 went on to receive a 4th dose of MeNZB, and 121 in the second group who received only the routine vaccine schedule.
As in earlier studies the immune response to MeNZB was measured by the % that achieved a ≥ 4 fold rise in their SBA titre 6 weeks after their 3rd dose, day 151, and the % that achieved a ≥2-fold rise. In the subgroup further blood sample was taken at

day 256, just prior to the 4th dose and at day 298, 6 weeks after the 4th dose. SBA GMTs were also measured at these times and the % of infants who demonstrated ≥ 4 fold increases of IgG antibody as measured by ELISA.
The immunogenicity tests against haemophilus influenzae type b anti-PRP, HbsAb, diphtheria and tetanus toxoids, 69K(Pertactin) and PT pertussis antigens were measured by ELISA. The immune response against Polio Types 1 – 3 was measured by a neutralising test.

Statistical methods:
Hypothesis was that at least a 95% confidence interval the proportion of responders showing at least a 4 fold increase in SBA at 6 weeks after the 3rd dose in comparison with base line, day1, is ≥ 40%. With 200 subjects able to be evaluated, the 80% power to obtain lower limit of 95% confidence ≥ 40%, assumed a true value of at least 50% of subjects are SBA responders.
Local and systemic reactions were recorded descriptively.
Results:
Results are given in **Module 5 Vol. 6A Tables 2-2 through to 2 –8,** attached.
Table 2-2 shows that 53% (CI 46 – 59%) of Group1 achieved ≥ 4 fold rise in SBA titre, but 0% in Group2.
Table 2-3 shows that 76% of Group 1 had a titre of ≥ 1: 4 and 54% a titre of ≥ 1: 8 by day 151. 98% had shown a 4-fold increase in ELISA by day 151
Table 2-4 shows that in the 57 infant subset, the % of ≥ 4 fold SBA responders at day 151 was 48% but had fallen to 13% just before the 4th dose. There was a further rise in 4-fold responders to 69% following a 4th dose of vaccine. 82% of the infants had achieved a ≥ 2-fold response after the 4th dose and 82% had a titre of ≥ 1: 4 and 69% ≥ 1:8.. The GMT of SBA was 22. 100% showed ≥ 4fold rise in ELISA titre and the GMC was 159.
Table 2-5 shows that both those who received MeNZB + routine vaccines and those who received routine vaccines alone achieved satisfactory immune responses to the routine vaccines after 3 doses with some evidence that the concomitant administration with MeNZB increased the response to all but the Hepatitis B. Minor differences were noted in the PT + Pertectin resposes.
Note: The Hepatitis B response figures have been amended to show that 78% in the group who received both MeNZB and routine vaccines achieved a titre of ≥10 IU /ml while those who received routine vaccines alone had a ≥ 10 IU/ml in 82%. **See Table dated 15th June 2006:**
Tables 2-6 and 2-7 show that the concomitant administration of MeNZB and routine vaccines increases both local and systemic reactions to some extent but the differences were not severe or prolonged. There is no fall-off in reactivity between the 1st and 4th dose of MeNZB. **In Table 2-8** an increase in systemic symptoms (a transient rise in fever) and administration site reactions is noted in those who received MeNZB as well. These were almost all local reactions related to MeNZB.
Comment.
The immune response to MeNZB in young infants is of a lesser order than in older children and adults and tends to be short lived. A 4th dose of vaccine induces a further response but its duration remains uncertain. The routine vaccines of the NZ immunisation schedule are not significantly inhibited when given with MeNZB, with the possible exception of HepBs antibody, and some may even be potentiated. This study does not show if there is any inhibitory effect in the response to MeNZB although the brisk response to the 4th dose does not suggest that there is. Local and systemic reactions are frequent but brief and not severe.

Vol. 7: Study V60 P4: (Final report dated 8th October 2004 – no update expected. See also the extension Study V60 P4 E1 below)
A Phase 1 / 2 open label single centre study to evaluate the safety, reactogenicity and immune response to 3 doses of MeNZB® when administered to healthy laboratory workers at ESR. This study has been previously reviewed. 10 subjects included. 90% had shown ≥4 fold rise in SBA 3 weeks after 3rd dose but 100% had SBA titre ≥ 1: 4 One subject failed to respond after 2nd dose but did so with 3rd while another subject responded after 2 doses but no had further rise with the 3rd. SBA GMT for the whole group was 2.46 at baseline and 33 (CI 9.41-113) 3 weeks after 3rd dose. 80% had ≥ 4 fold rise in ELISA after 3 doses. Adverse reactions were mostly local pain and headache but one suffered prolonged myalgia.

Vol. 8: Study V60 P1E1 (Final report dated 28th February 2005 – new study)
Extension of original Phase 1 / 2 study in which adult subjects were divided into 3 groups, two of which were given either 25µg or 50µg of MeNZB and the 3rd group given 25µg of MenBVac. The purpose of this study was to evaluate the persistence of SBA levels in the first two groups 10, 16 and 22 months following the 3rd dose. The results showed that there was a significant drop in SBA GMT from 27 one month after the 3rd dose to 6.01 (CI 2.88 –13) in Group 1 and 6.48 in Group 2 at 10 months, but minimal change at 16 and 22 months and in fact seemed to increase to 9.91 (CI 4.09 – 24) – **see Module 5 Vol. 8A Table 2.1-2.** The results of the ELISA titre showed a similar pattern.
In a further extension 13 subjects in Group 3, who had received MenBVac in the earlier study were now offered immunisation with MeNZB 10 months after their initial schedule. 15% had shown a ≥ 4-fold rise in SBA titre since base line of the original study, due to either a heterologous response or possibly to exposure to the epidemic strain of meningococcus B. After one dose of MeNZB 38% were now showing a ≥ 4 fold increase from baseline but 2nd and 3rd doses induced no further increases – **see Vol. 8A Table 1.1-3. In Tables 2.1-4, 2.1-5 and 2.1-6** it is shown that there was a moderate increase in the % of those showing a response of subjects with SBA titre ≥ 1: 4 and a rise in SBA GMT after one dose of MeNZB but little change with subsequent doses. A similar pattern is shown with the ELISA titres.
Comment: These results suggest that in adults who have responded initially either as a result of a primary response to MeNZB® or as a heterologous response to a closely related vaccine, there is no great advantage in offering a booster dose after the initial 3-dose course.

Module 5: Vol. 9A Study V60P3E1 (Previously reviewed).
A Phase 2 follow-up study, to evaluate the persistence of the immune response in subjects who received 3 doses of MeNZB during studies V60 P2 (aged 8 – 12 years at primary immunisation), V60 P3 (aged 16 – 24 months) and V60 P5 (aged 6 – 8 months).
All subjects have been reanalysed 4 – 14 months after completion of their primary immunising course. To allow for any variation in laboratory methods, samples taken one month after the 3rd dose of the primary course were reanalysed at the same time as new serum samples taken 4 – 14 months later. Samples were tested at both 4 & 14 months in the 8 – 12 year olds, but not in the same subjects.
Results are shown in **Module 5, Vol. 9A Tables 2.1-2 through to 2.1-6.**
In **Table 2.1-2** it is shown that SBA GMT in the 6-8 month infants has fallen from 27, one month after the 3rd dose to 2.36 at 7 months. The toddlers aged 16 – 24 months had decreased their GMT from 24 at 1 month to 1.76 at 11 months. The school age children from Cohorts A & B of Study V60 P2 (**see Table Module 5 Vol. 9A, 2,1-5.**)

had decreased from 18 at 1 month to 4.27 at 4 months and 3.11 at 14 months. Each group of subjects had shown an equally marked drop in the % of those showing a ≥ 4-fold increase in SBA from baseline. The greatest drop occurred in the 16 – 24 month old toddlers **(Module 5 Vol 9A Table 2.1-3)** from 98% at 1month post the 3rd dose to 6% at 11 months. **Module 5 Vol 9ATable 2.1-5** shows a similar drop in the % of those with a SBA titre ≥ 1:4.and ELISA titres **(Module 5 Vol. 9ATable 2.1-6).**
Comment: The evidence is that primary immunity following a 3-dose schedule of MeNZB may be quite short lasting, particularly in infants and younger children. The duration of immunity following a 4th booster dose is uncertain.(See Study V60P3 E2)

Module 5 Vol. 10 A: Study V60P3E 2 (Final report dated 17th March 2005)
This is a follow-on study brought about by the results noted in V60P3E1, in which the tolerance of and immune response to a 4th dose in 2 groups of toddlers, one group of which had failed to show a response with ≥ 4 fold increase in SBA to the primary schedule of 3 doses of MeNZB. This group received a 4th dose at 11 months from Day 1 of the primary course. Others who responded initially but had lost their immunity were offered a 4th dose at 14 months. Vaccine Lot 030101, expiry date 31.10.04. used in this study.
Module 5 Vol.10ATables Module 5 Vol. 10A 2.1-2, to 2.1-6 tabulate the results
Of the 30 toddlers who failed to respond to the initial immunising course, 100% showed a ≥4-fold increase in SBA 4 to 6 weeks after the 4th dose. Similarly those whose immunity had waned responded 100% to a 4th dose. **(See Table Module 5 Vol.10A 2.1-2).** None of the first group had shown any response initially but 32% of the second group had SBA titre responses of ≥ 2 fold before the 4th dose. Both groups achieved a SBA titre of ≥1:4. GMT levels rose to 69 after the 4th dose in the initial non-responders and to very high levels of 259 in those with an initial response, which waned. **(See Module 5 Vol 10A Table 2.1-5).** An IgG based response, after primary immunisation, in both groups of toddlers as shown by ELISA.
(Module 5 Vol.10A table 2.1-6). 53% of the toddlers in the 1st group given a 4th dose at 11 months and 49% of those who received the 4th dose at 16 months suffered one or more adverse reaction following the extra dose but most were mild and none severe.
Comment: This study demonstrated that children re-immunised with a 4th dose were primed by the initial 3 doses, whether or not they had responded with a rise in SBA titre. A 4th dose gave rise to a marked immune response in each case and was the rationale for offering a 4th dose to children under 3 years. The duration of this increased level of immunity is uncertain.

Module 5: Vol. 11: Study V60P4E1: (Final Report dated 5th December 2005)
This is an extension of the original Phase 2 study in adults working as laboratory scientists at ESR who had received a primary immunising course of 3 doses and were at occupational risk from N.meningitidis B disease. The intention was to study their level of immunity 15 months after their 3rd dose and the kinetics of the response in individuals following a fourth dose. Six staff members were recruited to the study. Blood samples were taken prior to the 4th dose and 12, 24, and 48 hours on Day 5 and Day 8 following the extra dose, to determine the onset of an immune boost as measured by ≥ 4fold rise in SBA and by a rise in IgG titre by ELISA. Vaccine lot 040801A Expiry date 30.10.05 used in this study.
Module 5 Vol.11 Table 5.3.5.2.5 CSR shows that all 6 subjects had shown some response to the initial 3 dose vaccine schedule although in subject 01/006 it was at a low level while that of subject 01/001 the response was very high. All subjects had shown a fall in SBA titres 10 months after the 3rd dose immediately, before the 4th

dose, although it was not as marked in subjects 01/004 and 01/009. **The graphs** of response show that in each case a rise in the SBA titres did not occur until the 8th day after the 4th dose of vaccine. By Day 5 none had achieved a ≥4-fold rise in SBA. As noted in the graph the final levels achieved after the 4th dose were similar to those reached after the 3rd. Only 1 subject showed a 4-fold rise in ELISA.
(**Table Module 5 Vol.11 2.1-2**) and 2 subjects a 4-fold rise in SBA by Day 14. Two had shown a 2-fold rise by Day 7 (**Vol.11 Table 2.1-4**) and all had a SBA titre of ≥1:4 by Day 14, (**Module 5 Vol.11 Table 2.1-5**) 5 had retained a titre at this level 15 months after their 3rd dose. Rises in SBA GMT and ELISA GMCs were equally sluggish, not doubling from baseline until Day 7 for the SBA GMT.
Comment: This study indicates that SBA and IgG antibody levels against the epidemic strain of N.meningitidis B fall over time, even in adults who have shown an initial satisfactory response and that any booster effect from an extra dose or natural infection will be sluggish, stressing the need to maintain a level of immunity in the community while the present epidemic strain persists.

Module 5: Vol. 12: Study VA 00-02(I62P1) (NIPH Study dated January 2004)
This was a double blind, randomised, Phase 2 study carried out in teenagers by NIPH to test whether the MenBVac, produced in a new production facility was equal in its immune and safety when compared with the vaccine produced in its previous production centre. 374 students from Oslo secondary schools were randomised in a ratio of 2:1 to receive MenBVac 25µg or placebo, in two doses, at 6 week intervals, extended to a 3rd and in some a 4th dose (given at 13 months after the 3rd). (Results of 4th dose not reported).
Following the 3rd dose ≥ 4-fold increase in SBA was noted in 75%, a level not inferior to that achieved by the previous vaccine used during the Norwegian epidemic. The new production vaccine differed in not containing thiomersol. Adverse reactions were similar to previous studies.

Module 5: Vol. 13: Study CSR VA 98-03 (I55P1) (Reported January 2004 – final data due February 2006 but not contained in the dossier)
A single blinded, randomised & prospective study in healthy adults to investigate the immune response and adverse reactions following delivery of MenBVac® and Menujugate® (Menigococcus C vaccine) either in combination (Group 1), separately (Group 2) or Menjugate alone. $Al(OH)_3$ was given as a placebo to make up the second and 3rd injections in Group3. Samples for SBA and ELISA serology were taken at 6, 12, and 18 weeks (6 weeks after each dose of vaccine) and at 1 year.
Results showed that at baseline Day 0, the SBA GMT for MenB was higher in those who received the vaccines combined than in those who received them by separate injections (4.76 to 2.89 - a ratio of 1.64). By 18 weeks the combined vaccine showed a greater difference in response (SBA GMT 29 to14 - a ratio of 2.01). At one year the ratio of difference was 2.25 (SBA GMT 16 to 7.29). The results were similar when ≥ 4 fold rise in SBA were compared, 59% at 18 weeks vs. 50%. As expected none of those receiving Menjugate alone showed a ≥ 4-fold rise against the Norwegian N.meninigitidis B epidemic strain,44/76. At 1 year the figures were 52%, 40% and 6% respectively.
For the Menjugate vaccine the SBA GMT MenC was 7.34 at Day 0, at week 6 it was 100 in those receiving the combined vaccine and slightly higher at 120 where Menjugate was given by a separate injection. At one year it was 39 for the combined vaccine group, 44 for the separate group. The proportion achieving ≥ 4-fold rise in MenC SBA was 69% and 68% at 6 weeks and 48% and 47% at one year.

ELISA results suggested that the combined vaccines rather than the separate regime was slightly more immunogenic against MenB – 97% versus 93% at 18 weeks and 74% versus 60% at one year maintaining a ≥ 4 fold rise. There was no significant difference in the ELISA results for the MenC antibodies at 6 weeks, 90% and 89%. At one year the difference was 35% and 41%.
Local pain and reaction was more prominent in those who received MenBVac either alone or in combination.
Comment: Combining a meningococcus C vaccine with a meningococcus B has been shown to enhance the response to the Group B without inhibiting the response of the Group C. vaccine significantly.

Module 5: Vol. 14. Summarises the 25 studies carried out with the Norwegian vaccine MenBVac®
The volume includes a description of the early development of MenBVac and decision to remove thiomersal at the request of the European Committee of Proprietary Medicine Products. Earlier vaccines also included Group C polysaccharide vaccine but was later removed because of significant difference in the Group B response and because of possible confusion in Group C specific diagnosis. Initial decisions settled on 2 doses 6 weeks apart, but when immunity had declined by 29 weeks a booster dose was given at 9 – 10 months. Later a 3rd dose was added, after an interval of 7 years from the 2nd,in a small group of 10 subjects who demonstrated antigen specific T cell immune responses with antibodies in the IgG 1 and IgG 3 subclasses and opsonophagocytic activity increased. A further study confirmed that the 3rd dose induced strong antigen specific T cell responses with an increase in the proportion of memory T-helper cells. Other studies showed that giving the 3rd dose 2 months or 9 months after the 2nd did not induce major differences in the response.
18 of the clinical trials with MenBVac were carried out in Norway, others in Iceland Chile and Cuba.
In a major double blind placebo controlled Norwegian clinical trial which involved 179,657 students in 1335 schools, 36 cases of proven acute severe Group B meningococcal disease occurred at least 14 days after 2nd dose of vaccine. 12 of these occurred in 11 of the 690 schools where the students had received MenBVac and 24 cases in the 645 schools where they had received a placebo. Most of the vaccine breakthroughs occurred more than 30 months after the 2nd dose. The estimated protection rate over 29 months was 57.5% -(p value 0.012, lower CI 28%). For a shorter time interval of 0.5 to 10 months the protection was estimated at 87% (CI 8-98%).
In a further study 2 cases of serious neurological disease, one of Guillain Barre′ disease and one of Mononucleosis with acute cerebellar syndrome, occurred following MenBVac, 53,037 subjects receiving at least 1 dose, 49,188 going on to receive dose 2nd. Two cases of Guillain Barre′ occurred in the placebo group.
Comment A separate report on the association between Meningococcus B vaccine and neurological complications has been circulated, which did not suggest that there was an increased risk from immunisation with MenBVac.

The rest of this summary report deals with various immune response and adverse reaction trials in military recruits, adults, infants and children already circulated.
One study showed that 46% of those who had been immunised with 2 doses of MenBVac remained pharyngeal carriers of N. Meningitidis, compared with 58% in those who had not received the vaccine. Most of the isolates were non-typable so that there clear evidence whether or not vaccination will reduce the frequency of pharyngeal carriage of an epidemic strain.

Module 5: Vol. 15A: Study V60P7 (Final report dated 15th December 2005)
This was a two part clinical study carries out in students living in a single hall of residence at the University of Otago, Dunedin.
Part A.: To Evaluate Meningococcal Carriage Rates in University Students.
Part B: A Phase 1 / 2 Observer Blind, Randomised, Single Centre, Controlled Study to Evaluate the Immune Response, Tolerability and Safety of Three Doses of Chiron MeNZB, Administered Separately or in Combination with a Single Dose of Chiron's Meningococcal Group C Conjugate Vaccine (Men C) in Healthy University Students
Methods:
In Part A, 209 of the 330 students were enrolled, were questioned about possible risk factors including smoking, alcohol consumption and number of visits to a hotel bar. Throat swabs were taken at the beginning of the study and then again approximately 5 months later, at the end of a semester, at which the same social functioning questions were asked.
In Part B 75 students who were a subset of those in Part A and were also questioned and had throat swabs taken, were randomised into two subgroups in a ratio of 2 / 1. The 38 in the first group were given 3 doses of vaccine in a schedule of MeNZB 1st dose, a 2nd dose of MeNZB / MenC in which the MenC was reconstituted with the liquid MeNZB and a 3rd dose of MeNZB at 6 week intervals. The second group received 3 doses of MeNZB alone. Blood samples were taken for SBA and ELISA titres before the 1st dose of vaccine, Day 13 of the study – before immunisation, 6 weeks after the 2nd dose and 4 weeks after the 3rd. Additional pharyngeal swabs were taken at Day 13, at Day 97, 6 weeks after the end dose of vaccine and Day 167, approximately 5 months after Day 1 Group B serology was carried out by ESR, as was the molecular typing of isolates. Chiron, at Marburg in Germany, undertook group C serology.
Exclusions were similar to those in previous studies.
Results:
Results of the studies are summarised in **Tables Module 5 Vol. 15A 2.1-2 to 2.1-15.**
In Group B, the response of ≥ 4 fold rise in SBA against the NZ98/254 strain was similar by 4 weeks after the 3rd dose in those who received the combined vaccine and those who were given MeNZB alone- 68% and 67% **Module 5 Vol 15A table 2.1-2)**
The response was greater in Group 1 when measured by the % subjects who achieved a type B SBA titre of ≥ 1:8 **(Module 5 Vol 15ATable 2.1-3).** 85% and 72% achieved a two-fold increase in Type B SBA **(Module 5 Vol.15ATable 2.1-4).** SBA GMTs were higher in those who had received the combined vaccine **(Vol.15A Table 2.1-5)** but ELISA responses were similar in both groups **(Vol.15 A Table 2.1-6).** Absolute titres for SBA against the NZ98/254 strain in **(Vol 15ATables 2.1-7 and 2.1-8)** and were similar for both groups although the combined vaccine recipients showed slightly higher responses.
Vol 15A Table2.1-9) shows that there was a brisk GMT response to the MenC vaccine in the combined vaccine group and a slight rise in the others who had not received MenC. The baseline titres were similar in both groups suggesting that contact with Group C meningococcus is relatively common.
(Vol 15A Tables 2.1-10 to 2.1-14) record the results of the pharyngeal swab analyses. The over all carriage rates for N.meningitidis was between 20 and 40% with only 2% harbouring the NZ epidemic strain. This did not vary over all from the beginning to the end of the study although individuals showed changes – **(Vol 15A Table 2.1-11).** One individual who received MeNZB had type B; 4:P1.7-2,4 at the first visit but lost it subsequently while another did not have the strain at visit 1, tested positive at visit 2 and then became negative again. Three of the non-vaccine recipients were carriers at the end of the study. A variety of other N.meningitidis strains were isolated from the

pharyngeal swabs (**Vol 15A Table 2.1-12**). **Vol 15A Table 2.1-13** gives the odds ratio for being a carrier of N, meningitides in both Groups A & B, at the beginning of the university term and **Table 2.1-14** the same list of ratios at the end. Although being a first year student, not receiving MeNZB, and alcohol intake had some influence on the risk of carriage, the only one which reached statistical significance was more visits to a Pub.
Vol. 15ATable 2.1-15 lists an overview of the reaction events associated with the vaccine and was similar to those seen in other studies.
Comment: This study was of interest as it demonstrated the relatively high rate of N.meningitidis carriage amongst young adults, also seen in some of the Norwegian studies, but very few were of the NZ epidemic strain. The greatest risk incurred was by going to a crowded public place, a Pub.

Module 5: Vol. 16 Clinical Study I60P1 (Report dated December 2005)
This was a Phase 2 study carried out with adults in the UK. Blood and saliva samples were to be taken at 6, 12, 18 weeks and 15 months, with a second blood sample to be taken 10 days after each dose. 13 subjects were recruited but only 9 completed. The CD file did not include any results.

Clinical Study I60P3 (Reported December 2005)
This study was designed to evaluate the mucosal immune response to 3 doses of MeNZB in adults and teenagers by taking samples 8 weeks and 2 weeks before tonsillectomy, on the day of operation and 4 weeks later. Again no results are included on the CD file.

Clinical Study I60P3 (Report dated January 2004)
This is a Chiron designed study in which recipients were to receive a vaccine made from 2 vaccine strains, MenBVac and MeNZB, 12.5µg of each antigen and 25µg of each vaccine separately. Serology was undertaken at Day 1, prior to immunisation and 6 weeks after each dose. A booster dose of vaccine, of a different strain from the original vaccines, was planned 12 months after the 3rd dose. Evaluation of the T cell response was also to be undertaken by measuring cytokine response following stimulation with meningococcal antigens.
The report outlines study methods and ethical procedures but no results included on the CD file.

Module 5 Vol. 17:
Final Report of the Independent Safety Monitoring Board and **Effectiveness Assessment**. These reports have already been circulated independently.

Independent Safety Monitoring Board Report:
The Safety Monitoring Board maintained a close surveillance throughout the immunisation programme and only sanctioned the expansion of numbers in each age group once it was assured that there had been no unexpected adverse events. The dates at which each expansion of the programme was sanctioned are reported. Screening for adverse events included the monitoring of infants and children under age 20 years either admitted to hospital or seen in Emergency Depts in 4 Northern Centres. Between 19th July 2004 and 27th November 2005 a total of 1,123,764 doses of MeNZB were administered. Specifically 402,651 doses - (187,252 1st doses) - were given to those aged 5 – 20 years in the period 19th July 2004 and April 17th 2005 (when hospital surveillance ceased for this age group). 317,711 doses have been given to infants and young children aged 6 weeks to 4 years between 19th July 2004 and 27

November 2005, - 115,694 their 1st dose, 106,543 their 2nd and 95,108 their 3rd dose. 33,062 doses were given to infants under 6 months of age in this period. Of the 65000 admissions to hospital or ED consultations in the 4 week to < 20 year age groups, in 3 monitoring hospitals, 47,000 were in the 5 – 20 year age group and 18000 in the 4 weeks to 4years. Of the 65000 admissions to hospital or ED, 3.734 (5.7%) met the criteria for possible vaccine related adverse events, and of these 2,242 (60%) met the case criteria, 1,191 (31.9%) met a clinical definition and only 63 (1.7%) met an epidemiological definition making the relationship with MeNZB probable.
In summary this report also indicates that there is no increase in the frequency of febrile seizures within 7 days following a dose of MeNZB in children under 5 years. There appears to be no statistical association between receiving a dose of MeNZB vaccine and either encephalopathy or acute flaccid paralysis, when a comparison is made with the rates of these conditions in children over the 5 years before the MeNZB vaccine was introduced.
There dose not appear to be any increased risk of a child developing Henoch-Schonlein Purpura or developing a recurrence of this condition following MeNZB immunisation and no relationship with an increased incidence of Kawasaki Disease.
The over-all rates of adverse events following MeNZB immunisation are low in comparison with several other routine vaccines.
There have been no deaths attributable to MeNZB immunisation and no increase in the frequency of infection related deaths in those who have received MeNZB.
A **Meningococcal B Vaccine (MeNZB®) Effectiveness Assessment** dated May 2006 reports a preliminary estimate of effectiveness of the vaccine in 0 –19 year olds who have received the full schedule of 3 doses of vaccine as 81%. 13 cases of vaccine break-through have been reported in fully immunised individuals, clinical infection occurring at least 28 days after the 3rd dose of vaccine. 7 of the 13 cases were in teenage subjects. There is some uncertainty about the effectiveness figures because it is thought that the epidemic may be waning naturally although a recent news media report of a drop of 76% in reported cases in South Auckland, since the vaccination campaign began suggests that the improvements are due to the introduction of MeNZB®

Module 5 Vol. 18: Epidemiology of Meningococcal Disease in NZ, 2002

Peak frequency of disease reached in 2001. Significant differences in age and ethnicity in different regions of the country although still highest rate in children <4 years in all areas. 82% of clinical cases were in those < 20 years. There was a relatively low incidence in the Central Health Districts North Island.
The highest frequency occurred in Pacific Island children < 5 years. Rates amongst Maori were generally lower in Midland, Central and Southern regions compared with the North.
83.6% of isolates in 2002 were Serogroup B. 92% of Serogroup B isolated were epidemic type P1.7b,4. Serogroup C increased in Otago in 2001, with a further increase in 2002 to 15.5% throughout the rest of NZ. 13 of 35 N.meningitidis isolates recorded in Otago wereGroup C. Similar pattern were demonstrated using PCR identification – 84% shown to have DNA encoding P1.7,4 PorA type. 6 of 16 deaths in which serotype identified due to Serotype C, 9 due to epidemic strain Serotype B and one unidentified.
Of 115 PCR positive cases from Otago, one had meningitis, 2 septicaemias, 12 a petechial/purpuric rash, 29 cases had clinical tonsillitis with aching joints, headache, stiff neck, photophobia, fever, and vomiting and flu-like symptoms. 21 of 72 were admitted to hospital. Most aged between 15 and 29 years. 3 were < 5years.

New Zealand wide 21.7% were given pre-hospital antibiotics – not much difference between the regions. Pre-hospital antibiotics more likely to be given in the 10 – 19 age groups – 33 – 36%. Giving antibiotics prior to hospital admission reduced mortality 2.5x, if seen by a doctor, 3.5x if not seen by a doctor.
Total cases recorded between 1991 & 2002, 4752 with 203 deaths. Case fatality rates 2002, 3.2%.
Hospitalisation data was 87% sensitive in monitoring frequency of disease.
75% of cases laboratory confirmed in 2002.
Mean age for clinical cases 2.9 years for Maori, 3.0 years for Pacific Island and 16.0 years for Europeans.
90% of isolates exhibit PorA protein P1b,4. The Por B protein, which determines serotype, has shown variation.

Comment: The clinical studies all record that there is a satisfactory immune response to MeNZB® vaccine in a majority of subjects after 3 doses, as measured by SBA and ELISA, but the level of response is lower in young children, especially young infants and the duration of apparent immunity may be short. A fourth dose leads to an increased immune response in those children who have failed to respond following the first schedule of 3 doses or who have lost their initial response 10 to 14 months later. Study V60P4E1 shows that a 4th dose in adults induces a limited response in those adults in whom the immunity was waning, but only brought titres up to levels similar to those just after a 3rd dose. The rate of response was sluggish, suggesting that exposure to natural infection in one whose immunity has waned, would not be sufficiently brisk to give complete protection.
In a study of adults, those who had a response to a related MenB vaccine, (See V60 P1E1), showed a heterologous antibody response against the NZ98/254 strain had only a moderate response when given further immunisation with MeNZB, and then only after the first dose.
The study in University Students, V60P7, has shown that carriage with a wide range of N.meningitidis B strains is common but not of the epidemic strain in this group. Immunisation does not seem to prevent carriage although the numbers are too small for significance. The greatest risk for exposure is to visit crowded public facilities.

MeNZB Full Consent Application,

Module 2:Volume 1:
Introduction:
2.2: An introductory statement describing nature of MeNZB® vaccine. $Al(OH)_3$ included in formulation both as adjuvant to boost immune response and also reduces endotoxin effect of lipopolysaccharide (LPS). Vaccine stated to induce both humoral and cellular immune responses to P1.7-2,4 strain of N. meningitides B. It may also induce some degree of heterologous immune response against other serogroup B subtypes.

2.3.S1: General information given on structure of the outer membrane vesicles, OMV, which make up the antigen element of the vaccine. PorA protein defines sero-subtype of Group B meningococcus. All meningococcus strains contain Class 2 (PorB2) and Class 3 (PorB3) proteins, which define serotype. NZ epidemic strain contains Class 3 protein and is serotype 4. Class 4 proteins are immunogenic but antibodies are neither bacteriocidal or opsonic. Class 5 proteins also occur in small amounts, their structure is not well defined but they are highly immunogenic, bacteriocidal and opsonogenic. Laboratory method differences can account for significant variation in the determination of MWt of OMV antigens but make up an important part of production analysis to ensure presence of all main proteins.

2.3.S2: Manufacture:
Manufacture of MeNZB® undertaken at 2 sites. Initial innoculum and harvest to OMV suspension at Pre-bulk Concentrate stage at Siena site and purification, release testing, filling and packaging isn carried out at the Rosia site.
Original seed lot prepared by NIPH from a clinical isolate, NZ 98/254, made in 1998 in Whangarei.
Procedures model on those undertaken by NIPH in production of Norwegian vaccine MenBVac but changes made to exclude any substances of animal origin in the initial culture medium and also to facilitate large scale production. Some changes in excipients
Production methods are described. Original cultures deactivated with deoxycholate (DOC). Sterility ensured, following separation of OMV from whole organism, by multiple filtration and culture of filtrate to ensure no contamination. Each phase governed by pre-tested Standard Operational Procedures (SOPs) which have been developed by a pre-production laboratory and are audited and signed off by designated personnel at each stage of production. Each batch of vaccine is tested by Western Blot to confirm consistency of Class 1 PorA , Class 3 PorB and LPS content. Class 5 protein not considered to be important for efficacy of MeNZB vaccine. (See later statement which suggests that presence of Class 5 protein greatly increases the bacteriocidal immune response).
Care is taken to exclude animal products from manufacturing process. Deoxycholate used in inactivation of N.meningitides B following initial culture sourced from non-European/North American countries.

2.3.3 S3 Characterization:
Membrane components preserved as close as possible to their native form to preserve bacterial outer membrane mosaic. LPS content necessary to stabilize OMV vesicles.

May also be important in inducing IgG related antibodies. Stability of vesicles is checked during manufacture by electron microscopy.
Characterization of MeNZB vaccine has been compared with Norwegian MenB Vac in 3 production development batches, one of which was used in initial Phase1 and 2 studies.. Class1 P4 and Class3 Serotype 4 proteins found to be specific to the NZ strain.

2.3.S5 Reference Standards and Materials:
Initial antigen batch chosen by Chiron was a purified OMV bulk produced by NIPH. Identification testing of antigen content carried out 3 times on different days by two operators working independently using SDS-Page, Coomassie staining and immune blotting. SDS-Page required to be comparable within 11% over the 6 tests.

2.3.S.6 Container Closure System:
Type of glass used in vials described, to meet Ph Eur. standards, method of filling to ensure sterility and subsequent testing, with controls outlined and audited, as in all other stages of manufacture, before product release.

2.3.S.7 Stability:
Stability of vaccine tested following NIPH protocol – 4 batches stored at 5° and 25°C for 12 months and 24 months by Chiron with no evidence of instability at recommended and accelerated stress temperatures. Long term storage of 36 months at 2° - 8°C are to be carried out. Consistency of stability has been shown, on testing, from batch to batch. The protocol for stability testing is according to "Testing for Biotechnicological/Biological Products CPMP/ICH/138/95

2.3.P1 – P8: Description and Composition of Drug Product & Efficacy:
This is as described in the Drug Information sheet to be included in the packaging. Described as meeting with UK and Ph Eur. standards.
Apart from the nature of the antigen it differs from the parent vaccine MenBVac by excluding 3% sucrose and including NaCl, as a tonicity modifying agent, and histidine as a pH buffering agent. Both of these constituents have been approved by Ph.Eur. The $Al(OH)_3$ content is licensed as part of the solvent in the UK. The LPS content is less than in the parent MenBVac. The comparative efficacy evaluation was based on dose/response models in mice, measured by ELISA, measuring changes in optical density against a 4 fold parameter logistic regression curve, plus measurement of OMV protein content by SDS-PAGE and Immune blotting. Confidence limits of potency were wide ranging, from 41 – 243%, compared with "master" preparation.

2.4 – 2.6 Non-clinical Overview and Summary of Studies:
Discusses development of vaccines and comparison with MenBVac. Comparisons made between preparations of MeNZB manufactured by NIPH and Chiron support 'bridging' of manufacturing methods changing from Norway to Italy.
Animal experiments in mice and rabbits show MeNZB induces acceptable level of local and systemic reactivity, similar to those seen with MenBvac in man.
Immunogenicity tested by ELISA shows consistency between NIPH and Chiron produced MeNZB. Some variation in SBA response, depending on source of complement used and also strain of test mice used. Testing against strains other than NZ98/254 in mice following immunization with MeNZB shows some degree of heterologous immunity.

Because of the high degree of immune response, not possible to create a dose/response curve in mice.
No studies of T cell response have been carried out with MeNZB. Not thought to be necessary as earlier studies had been undertaken, in Norway, with MenBVac which showed increase in production of T-cell related cytokines.
MeNZB thought unlikely to interact with pharmacological receptors – eg. adrenergic, serotonergic – so thought it unlikely that vaccine would alter pharmacodynamic profile of typical medications taken at the time of vaccination.
Guinea pigs given 10 times human dose by body weight, with no ill effects over 7 days.
Pyrogenic tests carried out in rabbits according to Ph Eur. standards by NIPH and USP/2CFR standards by Chiron. Even smallest human dose induced pyrogenic effect but passed test according to smallest pyrogenic dilution.
No anaphylactic reactions noted in animal studies. Risk in humans considered to be low.

Studies using large and repeated doses (8) in pregnant rabbits show no increase in foetal loss or abnormality in comparison with non-immunised controls.
Animal studies included tests for residual deoxycholate (DOC) in blood following single dose vaccine. Measured at 0.002µg/ml. Levels in 15 healthy non-immunised human adults averaged 0.462µg/ml. A three dose vaccine regime expected to cause negligible increase in this level.
Because of its endotoxin activity, level of LPS content carefully monitored. Level set at 0.04 – 0.12µg/µg of protein to ensure both OMV stability and immunogenicity. Absence of LPS renders OMV unstable. Addition of $Al(OH)_3$ reduces pyrogenic effect of LPS to 2% of that induced by unbound LPS and overall pyrogenic effect 4 fold.

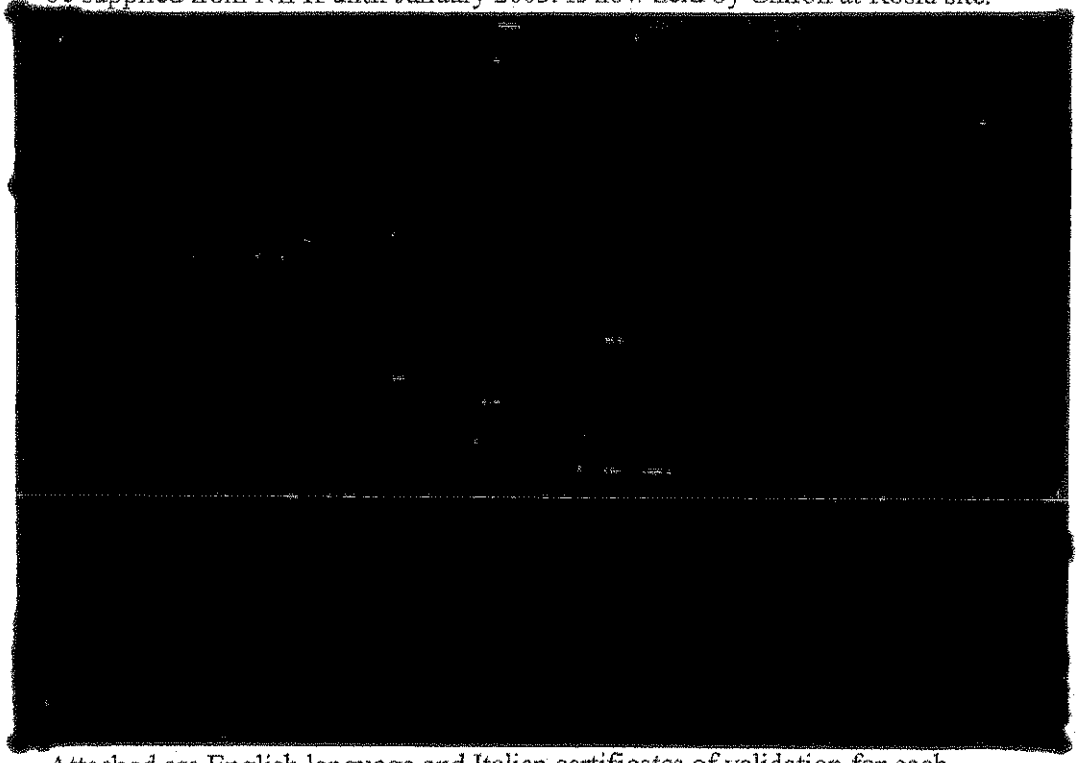
Discussion:
Human studies with Norwegian MenBVac in 250,00 recipients demonstrated an acceptable safety profile of the vaccine. Main adverse reactions included injection site tenderness, headache, nausea, general malaise and weakness. Main changes between MenBVac and MeNZB, apart from the specific NZ strain used in MeNZB, is in the production process where a specifically formulated synthetic fermentation medium, without material of animal origin, is used and 3% sucrose used in MenBvac is excluded. Histidine is used to control pH and NaCl to stabilize osmolarity. The similarity between the two vaccines suggests that their immune profile and toxicity will be very similar. This has been confirmed by clinical studies.

Module 3, Vol 1A Updated 4.1.2006
Contains a list of process validation and quality control updates, most dated December 2003 to December 2004..1 Endotoxin limit reduced from [redacted] to [redacted] August 2004.. Stability of vaccine updated to 24 months December 2005. Includes description of drug substance, [redacted]

Different analytical methods produce variations in recorded molecular weights, hence

need for strict consistency in both method and operator technique during batch analysis.

3.2.S.2.2 [redacted]. – Updated November 2004
Documentation also includes list of other vaccines produced at Rosia site, requiring close monitoring of cleaning sterilization and isolation procedures when changing production lines. Plans of production sites provided with indication of isolation procedures.
Original NZ 98/254 seed culture transferred from NIPH. Working seed continued to be supplied from NIPH until January 2005. Is now held by Chiron at Rosia site.



Attached are English language and Italian certificates of validation for each procedure.

Module 3 Vol 1B: Individual Process Validation Master Plans (PVMP)
Individual Process Validation Master Plans developed for Siena and Rosia sites are described. Effectively these are specific protocols for all stages of production and quality control and ensure that there is documented evidence of a high degree of confidence that each department, including all of individual areas, equipment and services, consistently give rise to consistent and reproducible results. This is carried out via the development, execution and documentation of all activities, entailing calibration, installation qualification, operating qualification, cleaning validation and qualification of all areas, equipment and services used for performing the production activities, and includes validation of the analytical methods – and further validation of the validation methods.

Facilities are in compliance with cGMP and subject to regular inspection. Because other vaccines are produced at the same facilities any shared equipment, product specific cleaning validations were also prepared to avoid cross contamination.
PVMP at Siena site ensures compliance with European Community requirements.

The protocol for the Siena site meticulously defines each phase of production from seed culture, through fermentation, concentration, and inactivation with an explanation of the rationale for each step, based on experiments conducted during the pre-production development phase and confirmed during earlier manufacturing batches.
Acceptance criteria are defined, validated and signed off by specified personnel. – Italian and English language versions are included. A report shows that the first 4 batches at Siena, numbers 001 to 004, complied with all of the acceptance criteria. There was one variation of the production lots early in the series, lots 001 & 002, when it was noted that the LPS/protein ratio changed to 0.05 –0.15µg/µg protein in the purified bulk. Application was made to the Authorities (MedSafe) to allow this variation and apparently permission was given although I could find no written validation of this approval. Lots 003 and 004 complied with the new specifications. This process of validation is continued through all future production lots.

A series of studies were carried out to ensure inactivation of the post-fermentation bulk product, both before and after filtration, using deoxycholate. The studies showed that inactivation was immediate, possibly due to the simultaneous lowering of temperature of the bulk product during the process.
A cleaning Production Validation Report (PVR) confirmed that after the cleaning process, sanitisation and storage, no protein, cleaning agents, bioburden or endotoxin residues were found contaminating the . Validation was carried out on 3 lots, compared with a negative control.
The manufacturing processes were designed to produce antigen sufficient for18000 doses per batch. The main process modification, from those used at NIPH, was to change to a chemically defined artificial Caitlin medium to avoid using substances of animal origin and to reduce iron content, which interferes with the filtration process. Soy based protein is used as a source of essential amino acids. The final product bulk antigen has been found commensurate with the NIPH, but has a slightly lower antigen content.

A similar series of Process Validation Reports are presented for the production stages carried out at the Rosia plant.
Because of a need to have the MeNZB vaccine available for the immunization campaign as soon as possible, many of these validation procedures were carried out using the last 3 batches produced during the Process Development laboratories and the product used in non-clinical toxicology trials and some clinical studies (V60P2 Cohort B) and V60P4. Tests carried out by a 2 operators, working independently and in duplicate, with no more than 5% variation permissible between tests.
Reference standards for appearance, antigen purity, antigen identity, DOC content DNA and LPS content have been derived from in-house testing. Those for activity, protein pattern, endo-toxin content, sterility and total protein are according to Ph. Eur. standards.
Particular attention is paid to the validation of the structure of the OMV and the characteristic of the [redacted]. [redacted] and [redacted] proteins not thought to be important for the MeNZB vaccine – Opa proteins vary markedly in sequencing. [redacted]

Tests carried out on both MeNZB and MenBVac vaccine antigens, for comparison, using SDS-PAGE gel, has demonstrated that Class 1,3,4,5 & LPS content show

similar patterns between the NIPH produced and Chiron produced MeNZB Repeated tests carried out on concentrated pre-sterilisation and post sterilization lots by 2 independent operators, in triplicate, on 3 different lots, showed good reproducibility and repeatability.
Rationale for the choice of antibodies used for Western Blot analysis of Class 1 P1,4 protein and Class 3 serotype 4 protein which are specific to the NZ98/254 strain and differ from those for Norwegian44/76, is given, whilst LPS 3,7, 9 and Opc antibodies are common to both and the same antibodies can be used.. Antibodies to other proteins considered not needed for test. (**See Module 3 Vol 1C**, below for further detail)

Validation of analytical procedures for endo-toxin content is critical for the safety of the product and to give an indication of its biological activity as 7-9% LPS content needed for OMV stability. A process, the Limulus Amilocyte Lysate (LAL) test has been carried out, according to the Ph. Eur. technical monograph, and has shown compliance in the 3 production development batches. The test repeatability has been confirmed and validity verified by deliberate "spiking" of some test specimens. With excess LPS.

Sterilisation of the product during production is carried out by membrane filtration. Tests have been carried out on the integrity of the membranes and their efficiency in removing any bacterial or mycotic bio –burden and efficiency has been confirmed. Similar validation of DOC content has been carried out using a method developed by NIPH.
A similar set of validation tests were carried out by NIPH on 3 MeNZB lots produced by them and showed similar consistency although an increase in the LPS/protein ratio was noted with the new production method, compared with their methods in producing MenB Vac.

Module 3 Vol 1C:
Reference standards and methods for identification and quantifcation of antigen content. Updated November 2005.
Purified bulk preparation produced by NIPH MeNZB, used in early clinical trials, chosen as standard for comparison of subsequent production lots. Later bridging process undertaken from the NIPH based to the Chiron based standard preparation. Class1 and Class 3 protein antibodies secured from NIPH and NIBSC in the United Kingdom. Some cross-reactivity, between different N. meningitidis B strains shown by NIBSC anti-sera so NIPH anti-sera chosen to verify specificity of Class 1 and Class 3 antigens. Antibody for Class 5 Opc obtained from both NIPH and NIBSC.
Attachment 3.2.S.5-1 is an affirmative qualification report comparing two batches of MeNZB antigen one prepared by former NIPH method and new Chiron process for purity and identity.
A similar qualification standard has been produced for the LPS content.

Validation of Sterility of Equipment and Stored Product:
The next three sections describe the validation of the alarmed refrigeration storage of the bulk OMV antigen at the Rosia site, sterility tests for the tryptic soy broth used during filtration and the efficiency of 2 autoclaves used for sterilization of equipment with acceptance criteria. A technical report verifies the continuing sterility of a quantity of bulk NZ OMV stored at 2° -8°C for 24 months.

Stability testing:
This section contains data related to 4 lots of MeNZB produced at NIPH and stored for 12 months and 3 Production Development lots plus one Manufacturing lot produced by Chiron and stored for 24 months. Batches of Bulk OMV have been stored at 25°C for 2 months and 2° - 8°C for 18-24 months with no evidence of instability. Batches of bulk antigen, prior to vaccine formulation, are tested for stability after real time storage at 2° - 8°C every 3 months in the first year after production, every 6 months in the 2nd year and then annually, together with testing of pH and protein content, to conform with the recognized Biotechnological/Biological Products CPMP/ICH/138/95 standards. Further testing is to be carried out on bulk OMV following storage at 23° - 25°C for 2 weeks, 1 & 2 months according to the NIPH protocol. Accelerated temperature studies will elucidate any degradation problems in the vaccine, at room temperature.
Results obtained with the first Chiron manufacturing batch confirms that all data still within specification limits after 2 months at 25°C and after 24 months at 2°- 8°C when sealed in a glass container.

Module 3 Vol. 2A: Drug Product, Storage & Stability testing (Modified 5.1.2006)
This section is largely a re-iteration of earlier statements. It describes the vaccine content, as in Data sheet and gives the rationale for the make up of vaccine, including excipients.
The first clinical studies carried out with NIPH produced MeNZB vaccine which contained 3% sucrose as a stabilizer and suspension agent. This is no longer included by Chiron in its MeNZB®, which has been used in the vaccine roll-out campaign.

Further stability studies have been carried out by Chiron in the US. with the whole vaccine stored in glass vials for up to 18 months at 2°-8°C and up to 7 weeks at 36° - 38°C, and in pre-filled syringes following storage at 36° - 38°C for 4 weeks, and for 12 months at 2°- 8°C. Three vaccine lots have shown high degree of stability (98%) after storage for 18 months. No difference in stability was noted whether the vaccine was stored in glass containers or syringes.

Results from comparison studies between NIPH produced and Chiron produced MeNZB show that they are comparable in immunogenicity, toxicity and stability. pH stability is very efficient ranging between 6.5 +/- 0.3 in these studies.
Immunogenicity studies carried out in 8 mice using two different formulations of the vaccine, NIPH MenBVac OMV and Chiron produced MenB AG 287, a recombinant vaccine, which had been variously stored at 37°C for 5 weeks and after 3 months, 6 months and 18 months at 2° - 8°C. The two different vaccines, either MenBVac alone or a covalent MenBVac/AG287 vaccine, were given to the mice in 3 doses, intraperitoneally, at intervals of 0, 21 and 35 days. Immune responses to the vaccines were tested by ELISA and SBA. The immune response showed no significant difference in the response after the vaccines had been stored at 37°C for 5 weeks or at 2°- 8° C for 18 months. The tests were repeated using a combination of MenB OMV with another vaccine, M10 and the M10 alone as a control. The combination OMV/M10 showed the best immune responses after storage.

Comment: The nature of the AG287 antigen is not made clear. It appears to have been derived from a heterologous N.meningitides B strain (MenB 1000), unrelated to either NZ 98/254 or the Norwegian strain 44/76. The N menngitidis B OMV type vaccines appear to be remarkably stable under adverse storage conditions.

Manufacturing Process Development and Certification
Methods and analysis are as described earlier in this report. The process follows [redacted].

In the production of MeMZB® a list of critical steps and intermediates with stated limits, alerts and validations is included – Standard Operational Procedures (SOPs). The final specifications include certificates of identity, immunogenicity, osmolarity, and percentage of OMV adsorption to [redacted] according to internal SOPs. LAL level testing was carried out according to the Ph Eur Monograph.
Sterility is tested in a samples of 40 tubes, prior to and after filling, by adding fluid Thioglycollate medium to 20 and Soybean/Casein Digest Medium to 20. The 1st set is incubated at 30° - 35°C and the 2nd set at 20° - 25°C for 14 days. All tubes tested are required to remain sterile. A further set of vials are deliberately 'spiked' with B.subtlis, Cl sporagens, Staph aureus and Pseudomonas aeroginosa bacteria or Candida albicans and Aspergillus niger and incubated at 30° - 35°C for 3 days or 20° - 25°C for 5 days respectively, to check the sensitivity of the sterility testing. At the end of the incubation period all spiked samples showed characteristic turbidity while the controls remained clear.
Testing for pyrogens is carried out according to protocol by giving graded doses IV to rabbits according to USP and Ph Eur specifications. The final test dose was 0.214µg protein/Kg animal weight. A dose of 0.375µg/Kg produced fever in one animal in this series.

Container Closure System:
The vaccine is enclosed in colourless glass containers with grey bromobutyl rubber stoppers, aluminium over-seals with flip-off tops, which meet Ph Eur standards. The vaccine has been shown to be stable in similar containers. (See above). The stoppers have been shown effective in preventing contamination, by culturing a filled vial in a bacterial suspension of tryptic soy broth at 30° - 35°C for 7 days. The vaccine was found to remain sterile. These tests have been repeated several times by Chiron using a number of different types of stopper. Validation reports are attached confirming the container and stopper meet the required standards. There is also validation of the protocol for washing, sterilisation, filling, stopper insertion and foil crimping devices.

Light Sensitivity:
Photosensitivity by UVB and UVA radiant energy and with visible light shows that Class 4 protein is sensitive to UV but not visible light. The other proteins remain stable. The vaccine is not photo-stable according to ICH guidelines, **a fact which should be included on the labelling.**

Vaccine Compatibility with Container:
No incompatibility of vaccine product with syringes or needles has been observed or thought likely to occur. The containers, "glass containers for pharmaceutical use, Neutral Type 1, meet Ph Eur. standards.

Batch Formulation and Antigen Content:
There is variation between the size of batches of MeNZB antigen , dependent on yield following ultra-centrifugation and filtering. Part of the proteins, especially vesicle aggregates may be retained by the filters so that protein components vary slightly from batch to batch. **This was noted in an addendum to the validation Protocol for the Formulation of the MeNZB OMV vaccine dated April 2004.** This required a reduction in volume of 3 different formulation bulks from 30, 60 and 96 litres to 25, 40 and 85 litres respectively. A concentration range for Drug Substance has set to assure a protein concentration of 50µg/ml in the final vaccine, as required to conform to the certified acceptance criteria.
A further series of validation reports, obtained from process controls and release testing performed on 3 batches of MeNZB® showed that they had all met acceptance criteria, and confirmed the manufacturing process at Rosia.
In one series of tests of filling procedures 5 out of 11690 3ml vials were found to contain a variety of bacteria after incubation, a contamination level of 0.09%, which was within the action limits criteria set by the SOPs for the procedure. Subsequently 3 tests showed no contamination in 35727 vials during filling. Overall contamination rate was 0.02%, a rate confirmed by a second series of filling machine tests. These results have been validated and signed off.

Process simulations carried out at both Siena and Rosia have confirmed that the manufacturing environment, equipment, media. personnel and operating techniques were microbiologically safe and conformed to Company Quality Assurance protocols. All simulations were carried out under "worst case" environments in terms of sterility conditions and operatating staff.

Shipping Validation:
Tests have been carried out, both with empty containers and after loading, to ensure that a temperature of 2°-8°C is maintained on exposure for 72 hours with an external temperature of 11°-19°C. Temperature probes are placed in "worst case" positions. Concurrent validations are undertaken in New Zealand.

Control of Excipients:
The water for injection, Sodium chloride and histidine buffer are prepared locally at the Rosia site and meet Ph Eur. Specifications. The $Al(OH)_3$ is produced by Chiron at its Marburg site and is similar to the preparation licensed in the UK. Testing for bio - burden and sterility is performed according to Ph Eur. protocols. A certificate of analysis is attached.

Validation of Immunogenicity of Vaccine Batch:
The dossier contains a certificate of approval, updated November 2005, with a summary protocol of production and testing of MeNZB vaccine, and outlining the analysis and qualification of content according to methods noted above. The justification of specifications has been based on data from NIPH's production of MenBVac, plus from published literature (attached) adjusted for the NZ strain 98/254.

The series of tests includes validation of analytical procedures. Efficacy of the vaccine is reliant on serological data but cannot always be relied on from batch to batch. With N. meningitidis B vaccines it is not possible to mount suitably powered clinical studies. The SBA analysis is more variable than ELISA, because of the use of

an animal model. ELISA immune results together with qualification of relevant OMV proteins by SDS-PAGE and identification of key antigen epitopes, by immunoblotting, provides sufficient degree of assurance of batch potency and likelihood of efficacy. Chiron confident that it has chosen the right combination of test methods to assure each batch's ability to induce appropriate antibodies in humans to protect against systemic meningococcal B diseases caused by the NZ strain. Validation has included assurance of precision of the accuracy of test results, the setting of confidence limits, and the stability and robustness of ELISA measurements. Tests were carried out against 3 pilot Roll Out batches. Little variation shown between batches so batch No. 03-01-01 has been chosen as the in-house standard for immunogenicity testing. Cut-ff value for ELISA has been set at >938 ELISA units/ml. Acceptance value set at ≥1131 Units/ml, later extended to a range of 1131 – 5406 Units/ml. A suitable range of confidence limits around mean potency is set at 41% - 243%.
Validation of all quality assurance procedures for potency are carried out, for each procedure in triplicate, by two independent operators, 18 determinations of 3 different samples at 3 different MeNZB ELISA antibody levels are measured against a standard optical density curve. The cumulative coefficient of variation between tests was 4% with a range of 3 –8%. Signed off validation reports and technical report, together with analysis of 5 batches produced by NIPH and 3 by Chiron at Rosia are attached to the dossier. Some batches produced earlier in the series had wider acceptance criteria.

Shelf Life and Stability – updated November 2005.
Shelf life and stability had to be agreed upon before data was available for the Chiron product. It was agreed to bridge data from the Norwegian MenB Vac, quoted earlier in this review and the NW, OMV and Chiron MenBvac. Chiron has tested its own MeNZB® stored in prefilled syringes and glass vials for 1 month at 36° - 38°C , 6 months at 23° - 27°C and 24 months at 2° - 8°C. One batch of MenBVac lost potency after 12 months at 25°C.
Completed data is now available for the first 3 manufacturing batches of Chiron MeNZB at these temperatures and time intervals. Stability parameters have remained the same when MeNZB® stored at 2°- 8° C and 36° - 38°C for 6 months. One batch failed a potency test for the SBA level, when a different strain of mice was used, but there has been no other sign of instability. As noted earlier, Class 4 proteins are sensitive to UV light but this class of protein is not thought to be important in the vaccine.

Comment: As with the other Meningitidis B vaccines, MeNZB® appears to be remarkably stable under adverse storage conditions. It is intended to carry stability testing out to 3 years with an additional 30 month time point. Future commercial batches will be stability tested at 6, 12, 24 and 36 months, and to include antigen integrity, degree of adsorption to adjuvant, potency, endotoxin content, pyrogen testing, pH and sterility.

Module 3 Vol 3, Facilities and Equipment.
This volume details the buildings, facilities and equipment at the Siena and Rosia sites. At the Siena site the [redacted]

[redacted]before transfer to the Rosia site where [redacted]

The documents describe the layout of the buildings at Siena, the GMP classification of each section and lining of each area to prevent contamination, temperature control, air flow control and illumination. Personnel access is restricted. Other vaccine products produced at this site include H.influenzae B vaccine and C. diphtheriae protein. Special precautions are taken to prevent cross contamination between vaccines.
Rosia is a multi-product filling facility. At product change over, equipment cleaning and room clearance procedures,to avoid mixing of products and contamination are described. A total of 12 bacterial vaccines and 11 viral vaccines are manufactured at the Rosia site. Only one lot of production occurs at one time.
The Master Seed lot for MeNZB is also held at the Rosia site.

Vol 3 also presents a number of attachments in which the origin and validation of culture medium requirements and excipients are certified.

Vol 4:
There is a group of 18 literature citations provided, related to the culture of N. meningitidis in a chemically defined media and the manufacture of other N meningitidis vaccines.

Module 4
Vol 1: Pharmacology studies in animal models:
This module reports on the preclinical studies carried out in animals and is concerned with the immune response. A number of them have been referred to earlier in this review. No specific non- clinical studies of secondary pharmacodynamics, safety pharmacology, or pharmacodynamic drug interactions have been carried out in animals.
Studies were carried out to find the most suitable animal model to create a dose/response curve against the NZ 98/254 antigen and comparisons have been made with the Norwegian MenB vaccine strain. Further experiments were carried out in animals to facilitate the transfer of the manufacturing process from NIPH to the Chiron facility in Italy.
Mice and rabbits proved to be the most satisfactory study animals. Rabbit studies included foetal development studies following multiple vaccine doses in pregnant rabbits, as described earlier in this review.
Serum bacteriocidal titres are calculated as the reciprocal of the serum dilutions, obtained from immunised animals, which kill 50% of N.meningitidis B organisms as estimated by colony count. The ELISA titre is calculated from absorbance values against a standard logistic regression curve. Comparisons for both values are measured against both high and low titre quality control sera. Controls are included to ensure that no bactericidal activity is due to the complement or serum alone.

Early NIPH studies with Norwegian strain showed good response after 2 doses vaccine initially but sharp falls in immunity at.12 months, especially in younger age groups. Genetic variatns of the Norwegian vaccine strain have also showed significant variations with the immune response. Absence of Class 5 protein in their vaccine has caused a significant fall in the immune response. Removing Class 4 protein may have increased the immune response.

Mouse experiments have shown increased response after 3 doses, compared with 2, but a 4th dose has not caused a further rise against the homologous MenB strain but has stimulated a greater response against heterologous strains as measured by ELISA.

Giving two vaccine types at once increases the immune response against MenB. Genetically different mouse strains show different degrees of immune response against the MeNZB vaccine. Vaccine given intra peritoneally is highly immunognic in mice in doses of 10µg - 2µg but there is a more variable response to dose of 0.5µg.

The dossier includes a technical report on the embryo-foetal toxicity study of multiple doses in pregnant rabbits given 3 doses of vaccine pre-mating and 5 doses during pregnancy. This has been commented on earlier in this report Doses given were either 6.25µg, 25µg or 50µg,.with $Al(OH)_3$ as a control. No adverse systemic effects were noted with the adult rabbits during life or at autopsy.. Some foetal abnormalities noted but not outside normal for the species.

Module 4 Vol 4:
Genotoxicity, and carcinogenicity studies not considered applicable to this vaccine. Local tolerance studies show inflammatory changes only, which settle in a short period of time

Module 4 Vol 5:
References from the literature related to preclinical and clinical studies with MenB type vaccines.

Comment:
The material contained in Modules 2, 3 and 4 indicates that extensive pre-production testing has been carried out prior to the production of MeNZB®. In production each stage is carried out within strict Standard Operating Procedures which are independently validated. Every effort has been made to ensure that the product is sterile, safe and has appropriate potency and meet the criteria set down by the Ph Eur and other international agencies.
The production sites seem to be a quality to meet GMP standards.

Recommendations for MeNZB Meningococcal Group B Vaccine Full Licensure

August 14th 2006

I failed to include a final recommendation for full licensure in my original report, for which I apologise to the Vaccine Sub-committee members.
I believe that the vaccine should be registered for a license to be available for distribution in New Zealand, but this does not mean that it will become part of the regular schedule, nor do I think it should be. Such a decision is outside of the mandate of this sub-committee in any case.
My reasons for recommending full licensure are as follows:

- The frequency of Type B meningococcal disease has fallen in northern parts of New Zealand since the introduction of the vaccine, and although there was some was some decrease before the vaccine campaign was started it has been more rapid since then.
- The full effectiveness of the vaccine remains a question of doubt because it has not been possible to carry out a controlled trial but it is noted that in 10 of the 13 breakthrough cases reported in Table 3 of the Data Management Group's May report, only one 13 year old has been left with a significant ongoing sequelae. Nocturnal enuresis and behavioural changes are to be expected in a 3 year old after admission to hospital with a serious illness. Information on residual disabilities is not available for 3 of the cases.
- Two deaths have recently been reported following full immunization, but in one case it is not known, at the time of writing if the infecting organism was the NZ epidemic strain of Meningococcus B. In all of the initial Phase 2 studies, with the exception of the laboratory staff in V60P4, there were individuals of all ages who were non or poor responders to 3 doses of vaccine. I have not seen any reports of the SBA titres of any of the breakthrough cases at the time of their infection. None of the cases reported would have been of an age to receive a 4th dose.
- I share concern regarding the decreased response to HepB vaccine, the reason for which is not clear but may be related to methodology.
- Although the epidemic of Type B meningococcal disease seems to be lessening in New Zealand we cannot be sure that this is so in 2006. Although the studies have been limited there is no evidence that vaccination will reduce the frequency of carriage of the epidemic strain. Similar earlier studies in Norway showed similar results.
- Review of the manufacturing processes carried out by Chiron have convinced me that they made every effort to produce an effective and safe vaccine which is unique to New Zealand. It is my belief that if full marketing rights are not granted Chiron is unlikely to continue holding stocks for further use in the event of a recrudescence of the epidemic. This does not mean that I think that the present campaign should be extended at this time.