Appendix 6: Explanations of other biases relating to zanamivir by trial identifier. [posted as supplied by author]

NAIA2005 (treatment of ILI, prevention of progression to influenza)

- The aim was to recruit 273 participants with symptoms of influenza. In practice, 22 centres that were set up did not recruit participants. 38 centres, 8 in Canada and 30 in the USA, recruited 220 participants (111 of whom subsequently had laboratory confirmed diagnosis of influenza). The study was stopped before full enrolment because the influenza season had ended. NAIA2005 CSR only_rdct.pdf p4, p27 (p45 of pdf).
- Primary and secondary outcomes were redefined to be consistent across NAIA2005 and NAIB2005. This is reported in the revised analysis plan NAIA2005RAP_rdct, page 5 section 6.1.

NAIA2006 (post-exposure prophylaxis)

- Recruitment was in 16 centres, 5 in Canada and 11 in the USA. 41 other centres were set up but did not recruit participants. The aim was to recruit 380 participants. Only 64 contact case participants were recruited and randomised. Only one participant developed laboratory confirmed influenza illness. NAIA2006 CSR wwra-proj_rdct.pdf p viii (p8 of pdf), p21 (p39 of pdf).

NAIA30015

- The Glaxo Wellcome Research and Development Worldwide Product Surveillance and Pharmacovigilance Department had access to an open copy of the randomisation code.
- No information was reported about similarity of intervention and placebo.

NAIA30028

- Randomisation list was not included.
- The primary endpoint was originally the time to alleviation of the main signs/symptoms of influenza but this was later adjusted to the time to alleviation of fever; other endpoints included, time to return to normal activities and incidence of complications.
- Two participants receiving zanamivir reported an adverse event that led to withdrawal from the study; neither event was classified as serious. One participant experienced a febrile convulsion lasting five minutes on Day 1; they had fully recovered on Day 2. The investigator considered the event to be possibly drug related. The second participant reported stomach ache on Day 1 that lasted 12 hours; they had fully recovered on Day 2 and the investigator considered the event to be probably drug related.

NAIA30031

- Some outcomes included in CSR but not protocol.
- 630 randomised to placebo, 661 to zanamivir); it is unclear whether such a difference is likely to have arisen by chance.
- Exclusion criteria differed from protocol: protocol stated that participants receiving antibiotics for bacterial respiratory tract infection would be excluded.

NAIA30034

- Protocol states several other secondary outcome measures; these modifications are not listed in the 'protocol amendment' section:
    - The total number of OTC medications used.
    - The proportion of randomised subjects who require a prescription medication.
    - The total number of prescription medications used.
    - The proportion of randomised subjects who have an unscheduled healthcare contact.
    - The total number of unscheduled healthcare contacts.

NAIA3005

- Protocol amendment: Statistical methods revised to allow for stratification of vaccinated participants.
- In the efficacy evaluations, nasal symptoms (nasal congestion, rhinorrhea) were changed to nasal congestion (blocked, runny nose). NAIA3005 CSR wwra-proj_rdct p5 (p28 of pdf).
- Safety and non-vaccinated populations were not defined in protocol

NAIAB2008

- Combined symptom analysis as a secondary endpoint was added to the final analysis plan (14 May 1996). NAIAB2008 RAP_rdct NAIAB2008 CSR only_rdct p38 (p55 of pdf)

- High number of protocol violations: Reasons for protocol deviations are listed in Table 5 and Appendix 8. There were 238 (19%) participants with protocol deviations, 77 (18%) in the placebo group, 86 (21%) in the zanamivir *bid* group and 75 (18%) in the zanamivir *qid* group. Of these participants, 2 patients had influenza for greater than 48 hours at entry, 70 had no evidence of feverishness, 19 had less than two (non-feverish) symptoms at entry, and 107 had post-treatment visits after Day 8. In addition, 77 participants had treatment deviations which were defined as missing doses for two or more days as recorded in the Diary Card by the patient. A summary of protocol deviations for the Intent-to-Treat population is shown below. None of these deviations resulted in the participants being excluded from the efficacy or safety analyses

NAIAB2009 (post exposure prophylaxis study, Europe)

- Recruitment was in 73 centres in Europe and US; 37 other centres were set up but did not recruit participants. The study aimed to recruit 840, but randomised 575 participants. NAIAB2009 CSR wwra-proj_rdct.pdf pviii (p8 of pdf), p22 (p41).

NAIB2005

- The original intention was for 273 participants (91 participants per treatment group) to be recruited but due to the relatively low incidence of influenza, this target was not achieved. NAIB2005 CSR only_rdct p26 (p44 of pdf).
- The protocol for this study was amended twice. Both of these amendments were implemented prior to the commencement of recruitment. As a result of both internal discussions and external discussions with regulatory authorities, the statistical methods employed were substantially different from those documented in the protocol. NAIB2005 CSR only_rdct p26 p4 (p22 of pdf).
- Cough and sore throat reported as major symptom in CSR, but not in protocol. NAIB2005 CSR only_rdct p36 (p54 of pdf).

- Discrepancy between protocol and CSR regarding evaluation of viral shedding NAIB2005 CSR only_rdct p40 (p58 of pdf). NAIB2005 protocol wwra-proj_rdct p18 (p28 of pdf).

NAIB2006

- ITT population was stated to comprise all randomised participants. However, participants were not included who had no diary card data from Days 1 to 5 or were withdrawn prematurely from study treatment. NAIB2006 CSR wwra-proj_rdct (p41 of pdf).
- Participants who were at risk of developing complications were excluded from the study, and the investigators did not list complications as an outcome. NAIB2006 CSR wwra-proj_rdct (p24 of pdf).
- Inconsistencies in the Protocol Summary, Study Plan, and Study Procedures (Protocol 2 Amendment) (p23 of pdf).
- Cough and sore throat listed as primary symptoms in CSR but not in protocol. NAIB2006 CSR wwra-proj_rdct (p27 of pdf).
- Investigator global assessment not listed in protocol, but listed in CSR. NAIB2006 CSR wwra-proj_rdct (p28 of pdf).

NAIB2007

- Due to logistic difficulties in receiving results of influenza diagnostic tests within 48 hours of influenza symptom onset, some participants were recruited on the basis of a clinical diagnosis of influenza. NAIB2007 CSR only_rdct p5 (p22 of pdf).
- Cough and sore throat reported as major symptoms in CSR, but not in protocol. NAIB2007 CSR only_rdct p34-35 (p50-51 of pdf).
- Protocol amendment 3 which necessitated revising the definition of serious adverse events and time line changes for reporting adverse events. Amendments were also made to definitions for primary and secondary efficacy parameters, and statistical analyses were modified. NAIB2007 CSR only_rdct p21 (p76 of pdf).
- Protocol amendment 4 was made to the age range included (13 to 65 years, 16 to 65 years and 18 to 65 years, respectively) and inclusion or exclusion of patients with asthma, to meet local regulatory and ethics committee requirements. NAIB2007 CSR only_rdct p21 (p76 of pdf).

NAIB3001

- The study was conducted by 22 investigators in three countries (Australia, New Zealand and South Africa), only 13 of these investigators recruited participants. NAIB3001 CSR only-2_rdct p30 (p50 of pdf).
- The protocol defined the Influenza Positive population as a subset of the Intent-to-Treat population. This was changed in the Data Analysis Plan to be a subset of the Safety population. For this study, both populations were the same so the change did not impact the results. NAIB3001 RAP_rdct p6 (p15 of pdf).

NAIB3002

Protocol amendment 01 was applicable to all of the investigators. It corrected typographical errors and inconsistencies and included further categories to be used to assess secondary complications of influenza NAIB3002 CSR only_rdct p5 (p27 of pdf)

NAI30008

- There was an increased rate of liver dysfunction within the intervention group NAI30008 tables_rdct Table 81 (p168 of pdf).

NAI30009

- The authors used a different method of allocation concealment in different countries. “In the U.S./Canada, a double-blind label containing the details of treatment was affixed to each subject's treatment. In Europe, the Principal Investigator was supplied with sealed envelopes for each treatment number containing details of treatment.” NAI3009_CSR_only-2_rdct p11 (p35 of pdf).

- 471 participants were randomised to each trial arm with 247 in placebo and 224 in zanamivir. All these participants were included in the ITT analysis for symptoms. However, data were presented only for those with no use of relief medications and included this in the efficacy conclusions; see table 21 for the full results on symptoms. NAI30009 tables-2_rdct (p33 of pdf).

- “Mean overall assessment Diary Card symptom score over post-treatment assessments on Study Days 2 to 5. Missing Diary Card data was replaced by the previous entry. Participants with no Diary Card data were excluded.” So it appears that diary card was included if all was present, but excluded for participants who did not complete it, or the most recent day’s report was used. NAI3009_CSR_only-2_rdct p14 (p38 pdf).

- The study used different rescue medications for different sites. “For US/Canadian sites, the following relief medications were provided: Dextromethorphan cough suppressant supplied as guaifenesin USP 100mg and dextromethorphan hydrobromide USP 15mg/5 mL. Junior strength acetaminophen caplets were supplied as 160mg/caplet. and acetaminophen regular strength caplets were supplied as 325mg/caplet. Children's acetaminophen suspension liquid was supplied as 160mg/5mL. For European/Israel sites, paracetamol and pholcodine cough mixture was sourced and supplied as relief medication”. This is noteworthy, given that one of the secondary outcomes was Time to alleviation of clinically significant symptoms of influenza and no use of relief medication. NAI3009_CSR_only-2_rdct p9 (p33 of pdf).

NAI30012

- “Subjects were assigned to study treatment in accordance with the randomisation schedule. An unblocked randomisation schedule was used, due to the unpredictable nature of influenza outbreaks and the likelihood that some centres would recruit small numbers of subjects. Each subject was independently randomised to one of the two treatments, with no stratification by centre.” The randomisation schedule was not presented. More participants in the placebo group had a high-risk medical condition compared with the zanamivir group, in both the Intent-to-Treat and Influenza Positive populations. nai30012_csr-rdct p29 (p33 of pdf).

- “In the U.S./South America and Canada (1999/2000), a double-blind label containing the treatment details was affixed to each subject's study materials. In Europe, ROW and Canada (2000/2001), the principal investigator was supplied with sealed envelopes for each treatment number containing details of treatment. It was the responsibility of the principal investigator

to ensure that these envelopes were stored safely, but were readily available to the relevant staff. The GSK monitor confirmed with the principal investigator where the envelopes were stored for the duration of the study." It is unclear why one method for allocation concealment was chosen in the U.S./South America and Canada (1999/2000) and another chosen for Europe, ROW and Canada (2000/2001) nai30012_csr-rdct p28 (p32 of pdf).

- "The safety population included all subjects randomised to treatment and who took at least one dose of study medication." Data were analysed for only 166 (placebo) and 191 (zanamivir) participants. One participants appears to be missing from the placebo group. nai30012_csr-rdct p61 (p65 of pdf).

- Protocol Amendment 11 (dated 24 Nov 2000) applied to all sites and clarified exclusion criteria to ensure that participants with severe persistent asthma were no longer recruited into the study. It is not clear why after commencing the trial, participants with asthma were excluded. It may have been due to risk of bronchospasm, but this was not clearly reported. nai30012_csr-rdct p23 (p27 of pdf).

- Subsequent to Protocol Amendment 11 (Section 5.2), participants with underlying airways disease (e.g., asthma, chronic obstructive pulmonary disease) were informed of the potential risk of bronchospasm when using zanamivir. The participant was instructed that should they experience any such reaction, they should discontinue treatment and contact the investigator or clinic immediately. nai30012_csr-rdct p31 (p35 of pdf).

- "There have been spontaneous adverse event reports of patients being treated with zanamivir who have experienced bronchospasm and/or decline in respiratory function which may be acute. Bronchospasm and dyspnoea have therefore been included as undesirable effects in the core safety information (CSI) for zanamivir. In addition the CSI contains a warning that there have been very rare reports of patients being treated for influenza who have experienced bronchospasm and/or a decline in respiratory function after the use of zanamivir, some of whom did not have any previous history of respiratory disease." nai30012_csr-rdct p83 (p87 of pdf).

- More participants in the placebo group had a high-risk medical condition compared with the zanamivir group, in both the Intent-to-Treat and Influenza Positive populations. nai30012_csr-rdct p66 (p70 of pdf).

- Two deaths were reported in this study. It was reported that neither event was considered by the investigator to be related to study drug. Case narratives for these participants are presented in Section 13: this entire section has been blanked out. nai30012_csr-rdct p81 (p85 of pdf).