

Antivirals in seasonal and pandemic influenza—future perspectives*

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Antiviral drugs continue to be an important option for the treatment of influenza disease and will likely be the only option during the early phases of pandemic. However, the limited number of drug classes licensed for treatment of influenza raises several issues, particularly in the face of drug resistance. Two classes of drugs are presently licensed for treatment of influenza, M2 and neuraminidase inhibitors. M2 inhibitors are currently not recommended for treatment of influenza because of widespread resistance and resistance to neuraminidase inhibitors has been observed during the past influenza seasonal outbreaks. Additional antiviral drugs with novel mechanisms of action are clearly needed

for the treatment of influenza. Fortunately, the landscape of drugs in early and advanced development has dramatically increased over the last 5 years. Drugs targeting viral functions such as attachment, entry/fusion, transcription, and polymerase and drugs targeting host factors affecting viral replication are currently in clinical trials. Examples of these novel antiviral drugs and the challenges for influenza antiviral drug development are discussed in this article.

Keywords Antivirals, drug development, influenza.

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Introduction

Antiviral drugs have a long and extensive history in the treatment and prevention of influenza disease.¹ While their effectiveness has been debated, the need for influenza antiviral drugs was clearly demonstrated during the recent 2009 H1N1 pandemic when vaccines were not available for initial waves of the outbreak.² Two classes of anti-influenza drugs, the M2 and the neuraminidase (NA) inhibitors, are currently approved in many countries and are the only approved influenza antiviral drugs in the United States.¹ Despite the availability of these drugs, new antivirals are needed to address emerging virus resistance and gaps in treatment options associated with current therapies.

One of the primary issues for existing influenza antivirals is the constant threat of the emergence of viruses resistant to the limited number of approved drugs. The M2 inhibitors are currently not recommended for use because almost all circulating seasonal A viruses, including H1N1pdm09, carry a S31N point mutation in the M2 gene that makes such influenza viruses resistant to both amantadine and rimantadine.^{3–6} This has resulted in a heavy reliance on the NA inhibitors, licensed oseltamivir (Tamiflu®) and zana-

mivir (Relenza®). However, there are well-characterized point mutations in the NA gene that are associated with reduced susceptibility to this class of drug. A single H275Y change in the NA of the seasonal H1N1 strain circulating from 2007 to 2009 was associated with reduced susceptibility to oseltamivir, and there have been recent reports of the equivalent H275Y mutation in H1N1pdm09 viruses.^{7–9} Because H1N1 viruses with the H275Y marker have now been isolated from patients with no known exposure to NA inhibitors, there is concern that circulating influenza viruses with this characteristic will increase in prevalence, further reducing treatment options to drugs which retain activity against these mutant strains. The lack of drug options with different mechanisms of action that could be used alone or in combination has magnified the risk of widespread antiviral resistance developing to the limited treatment options currently available.

Another issue with the NA inhibitors currently approved in the United States is that they are only FDA-approved for uncomplicated influenza infections. While these drugs are routinely used off-label for complicated infections, they are not approved for use in severely ill, hospitalized patients even though numerous observational studies have

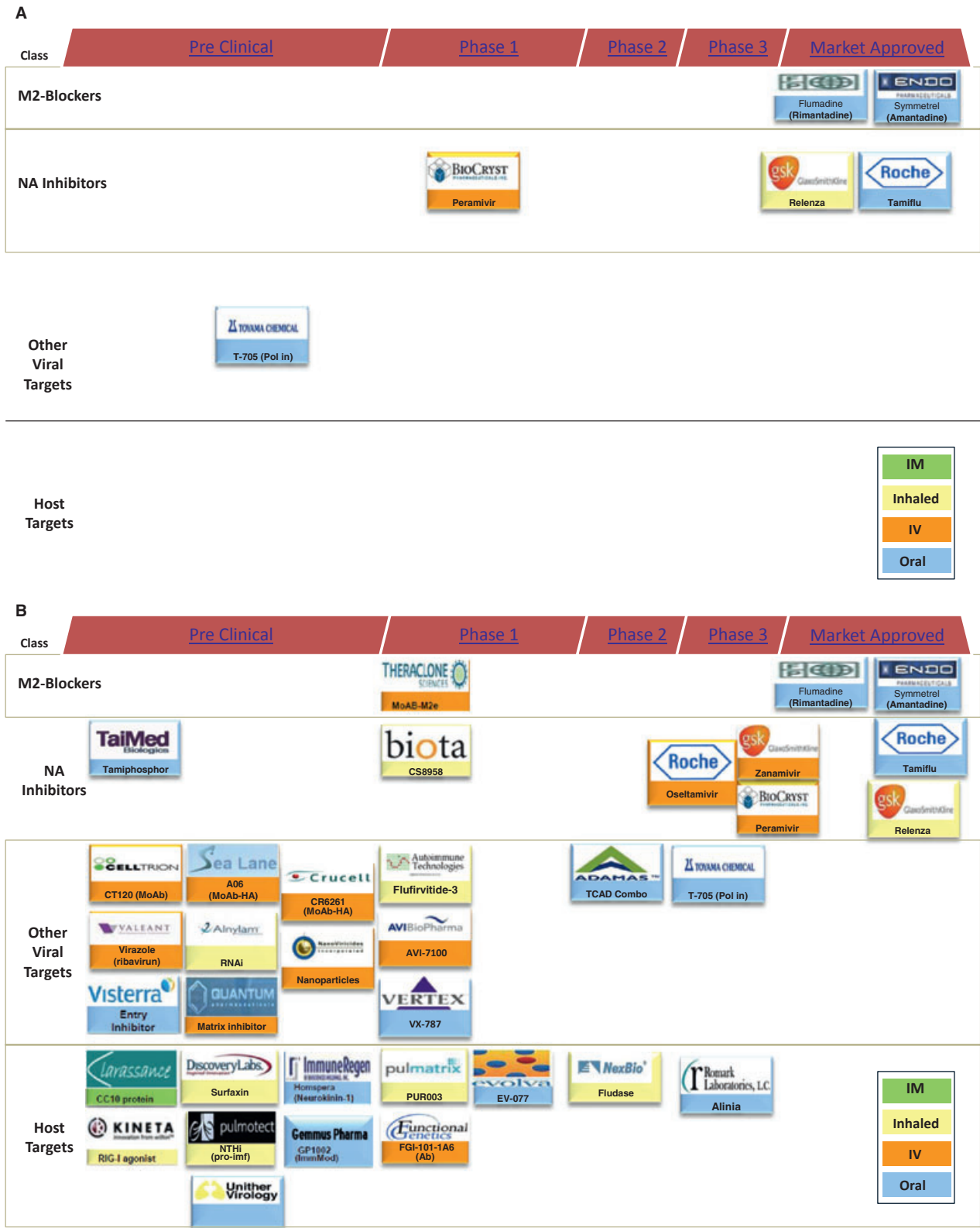


Figure 1. Influenza antiviral drugs approved in the United States or under development in 2006 (A) or 2011 (B). The compounds are indicated by name with the company sponsoring their development. Route of administration is indicated based on the color scheme shown in the legend. The drugs were categorized based on their mechanism of action and stage of development in the United States.

Table 1. Examples of novel influenza antiviral drugs under clinical development in United States

Name	Therapeutic entity	Mechanism of action	Development phase	Comments
Vertex VX-787	Small molecule	Not disclosed	Phase I	Novel mechanism of action (Non-M2 and Non-NAI)
AVIBioPharma AVI-7100	Modified oligonucleotide	Viral transcription	Phase I	20-mer phosphorodiamidate morpholino oligomer (PMO) IV formulation
Autoimmune Technologies Flufirvitide-3	Peptide	Entry/ Fusion	Phase I	16-mer peptide inhibits virus entry Inhalation delivery
Toyama T-705 (Favipiravir)	Small molecule	Polymerase inhibitor	Phase II	Nucleoside analog. NDA filed in Japan.
CruceCell CR6261/ CR8020	Monoclonal antibodies	Entry/ Fusion	Preclinical	Binds to conserved stalk region of HA, inhibiting fusion Group-specific spectrum of activity
NexBio Fludase	Recombinant protein	Attachment inhibitor	Phase II	Fusion protein combining amphiregulin and sialidase domains. Phase 2A study showed reduction in virus shedding.
Romark Nitazoxamide	Small molecule	Immuno- modulatory	Phase 2B/III	Approved for treatment of diarrhea caused by cryptosporidium or giardia Phase III influenza study showed 21 hour reduction in time to alleviation of symptoms
Evolve EV-077	Small molecule	Inhibition of prostanooids	Phase I	Dual thromboxane receptor antagonist and thromboxane synthase inhibitor Prevents virus inhibition of host immune response

shown improved outcomes including reduced mortality with the use of NA inhibitors.^{10–16} In addition, no formulation for intravenous (IV) delivery of NA inhibitors has been approved in the United States which limits treatment options for patients on ventilators. However, IV-delivered peramivir was used in the United States during the 2009 H1N1 pandemic under a federal Emergency Use Authorization (EUA)¹⁷, and IV formulations of zanamivir and oseltamivir were used on an eIND/compassionate basis. Pediatric patients are another population that can experience severe influenza disease yet remains underserved by the limited antiviral treatment options.

Based on the issues listed earlier, additional antiviral drugs with novel mechanisms of action are clearly needed for the treatment of influenza. Fortunately, the influenza virus life cycle provides a number of targets for antiviral intervention. Virus targets that are being explored include virus attachment, virus/cell membrane fusion and virus RNA replication.¹ In addition, influenza's utilization of host cell proteins for virus replication and its need to suppress the host's innate immunity provide opportunities to inhibit virus replication by targeting host functions associated with the virus replication cycle.^{18,19}

Influenza antiviral landscape

The landscape of drugs in early and advanced development in the United States for treatment of influenza has changed dramatically in the past 5 years (Figure 1). In 2006, two

M2 inhibitors (amantadine and rimantadine) and two NA inhibitors (oseltamivir and zanamivir) were licensed in the United States for treatment of influenza infections (Figure 1A). In addition, an IV-administered NA inhibitor (peramivir) was in phase I clinical development²⁰, and a virus polymerase inhibitor (T-705) was in preclinical development in the United States²¹ The paucity of new antivirals under development in the United States at that time clearly highlights our reliance on a limited number of therapeutic classes for treatment of influenza and the risk of losing those options due to development of antiviral resistance, as has been clearly observed in recent history.^{4,5}

In contrast, the influenza antiviral landscape in the United States looks very different in 2011 (Figure 1B). Phase III clinical programs are ongoing for IV formulations of three different NA inhibitors, potentially supporting the currently unmet medical need to treat severely ill, hospitalized patients. In addition, T-705 (favipiravir) has progressed to phase II clinical trials and a novel long-lasting NA inhibitor (laninamivir) is entering phase II studies in the United States.²² Furthermore, peramivir and laninamivir have been licensed in Japan and an NDA has been filed for favipiravir. Figure 1B also illustrates the plethora of new influenza antivirals with novel mechanisms of action in earlier stages of development in the United States These include therapeutics targeting viral proteins and others inhibiting virus replication by targeting host factors. Table 1 highlights some of these antivirals to illustrate the breadth of molecules and targets currently under development for treating influenza

infection and disease. These include a small molecule, an anti-sense oligonucleotide, a peptide and a monoclonal antibody inhibiting novel virus targets such as virus gene transcription (AVI-7100)²³ and virus entry/fusion (CR6261 and flufirvitide-3).^{24,25} Two examples of drugs in Table 1 affecting host factors illustrate different modes of inhibiting virus replication. EV-077 indirectly inhibits virus replication by inhibiting the increase of prostanooids associated with influenza virus infection.²⁶ Increased prostanooids impair the host's innate immune response, thus increasing virus replication. In contrast, nitazoxanide is believed to be an immunomodulatory agent directly stimulating the host's innate immune response.²⁷ Compounds targeting host factors also offer the potential for broad-spectrum antiviral activity. For instance, nitazoxanide which is currently licensed for the treatment of cryptosporidium or *Giardia* diarrhea, has been reported to inhibit adenovirus, coronavirus, respiratory syncytial virus and parainfluenza virus in cell culture assays.²⁰

Future perspectives in influenza antiviral drug development

One irrefutable prediction is that development of virus resistance to antiviral drugs will continue to drive the need for new influenza antivirals. The appearance of the H275Y NA mutation in seasonal H1N1 was a dramatic illustration of such vulnerability. Oseltamivir resistance in seasonal H1N1 viruses circulating in the United States shifted from <1% in the 2006/2007 season to >95% by the 2008/2009 Northern Hemisphere influenza season.⁹ This rapid rise in resistance to oseltamivir, associated with the H275Y change in the NA, seemed to occur in the absence of significant drug pressure, indicating greater transmission fitness of the variant. While the equivalent H275Y change in the NA of the H1N1pdm09 virus has not yet become prominent among currently circulating strains of influenza, there are indications of increased resistance, with some US states reporting >5% resistance during the 2010/2011 season. Importantly, an increasing percentage of resistant isolates have not been associated with NA inhibitor treatment.^{28,29} The appearance and increased prevalence of H275Y variants clearly demonstrates the potential for naturally occurring influenza viruses to become resistant to NA inhibitors. Therefore, the population as a whole cannot rely on newly circulating strains of influenza always being sensitive to oseltamivir or zanamivir.

Despite the medical need, market (profit) potential of influenza antivirals has limited interest in this area of drug development. Outside of Japan, the influenza antiviral market has been sporadic and unpredictable. Sales during most influenza seasons have been low, especially compared with other pharmaceutical products, whereas sales during the 2009 H1N1 pandemic increased dramatically and chal-

lenged production capacity. Challenges to more widespread use of influenza antivirals include the need to treat within 48 hours of symptom development and point-of-care diagnostic assays with high false-negative levels. These issues may act as a deterrent in physicians' prescribing practices.

Challenges in designing and conducting clinical trials also confound influenza antiviral drug development. As there are no accepted surrogate endpoints, efficacy is typically based on time to resolution of symptoms.³⁰ However, the number and severity of symptoms will vary on enrollment, thus often requiring large trial sizes to achieve statistically significant data on efficacy. The type of population targeted for the clinical trial can also dramatically affect enrollment. Clinical studies of uncomplicated influenza in outpatient populations can usually enroll large numbers of subjects relatively easily and can evaluate a new drug in a placebo-controlled study. However, the self-limiting nature of influenza disease in this population restricts the drug's potential antiviral effect. In contrast, clinical studies in severely ill, hospitalized patients are difficult to enroll, results can be complicated by co-morbidities and placebo-controlled studies are ethically unacceptable.³⁰

Despite challenges associated with the development of new influenza antiviral drugs, clearly unmet medical needs, rapid emergence of virus resistance to the limited drugs available, US government funding for advanced development of additional drugs and the 2009 H1N1 pandemic appear to have revitalized the field of influenza antiviral drug development. The US Department of Health and Human Services Biomedical Advanced Research and Development Authority has devoted over 450 million dollars to date to the advanced development of new antivirals to address unmet medical needs for treatment of severely ill, hospitalized patients and for long-lasting therapeutics.^{31,32} In addition, numerous novel therapeutic molecules with unique mechanisms of action are now in various stages of drug development. Successful development of these drugs to final regulatory approval will serve to broaden the arsenal for treating all age groups and special populations as well as to mitigate the risk of emerging resistance against any one antiviral therapy. In addition, it is important to evaluate the effectiveness of using influenza drugs in combination, a strategy successfully demonstrated with HIV antivirals for increasing efficacy and reducing development of resistance. Antiviral drugs targeting host factors may also offer advantages by reducing opportunities for the virus to develop resistance. In addition, drugs targeting host factors may offer broad-spectrum antiviral potential that could permit treatment of respiratory infections without first identifying the causative virus. In conclusion, the influenza antiviral development pipeline appears robust. These are encouraging times for influenza virologists and infectious disease clinicians.

Conflicts of interest

The authors have no conflicts of interest to declare.

References

- 1 Beigel J, Bray M. Current and future antiviral therapy of severe seasonal and avian influenza. *Antiviral Res* 2008; 78:91–102.
- 2 Yang Y, Sugimoto J, Halloran M *et al.* The transmissibility and control of pandemic influenza A (H1N1) virus. *Science* 2009; 326:729–733.
- 3 Abed Y, Goyette N, Boivin G. Generation and characterization of recombinant influenza A(H1N1) viruses harboring amantadine resistance mutations. *Antimicrob Agents Chemother* 2005; 49:556–559.
- 4 Bright R, Shay D, Shu B, Cox N, Klimov A. Adamantane resistance around influenza A viruses isolated early during the 2005–2006 influenza season in the United States. *JAMA* 2006; 8:891–894.
- 5 Bright R, Medina M, Xu X *et al.* Incidence of adamantane resistance among influenza A (H3N2) viruses isolated worldwide from 1994 to 2005: a cause for concern. *Lancet* 2005; 9492:1175–1181.
- 6 Nelson M, Simonsen L, Viboud C, Miller M, Holmes E. The origin and global emergence of adamantane resistant A(H3N2) influenza viruses. *Virology* 2009; 388:270–278.
- 7 Lackenby A, Hungnes O, Dudman S *et al.* Emergence of resistance to oseltamivir among influenza A(H1N1) viruses in Europe. *Eurosurveillance* 2008; 13:1–2.
- 8 Lackenby A, Gilad J, Pebody R *et al.* Continued emergence and changing epidemiology of oseltamivir-resistant influenza A(H1N1)2009 virus, United Kingdom, winter 2010/11. *Eurosurveillance* 2011; 16:1–6.
- 9 Sheu T, Deyde V, Okomo-Adhiambo M *et al.* Surveillance for neuraminidase inhibitor resistance among human influenza A and B viruses circulating worldwide from 2004 to 2008. *Antimicrob Agents Chemother* 2008; 52:3284–3292.
- 10 Louie JK, Yang S, Acosta M *et al.* Treatment with neuraminidase inhibitors for critically ill patients with influenza A(H1N1)pdm09. *Clin Infect Dis* 2012; 55:1198–1204.
- 11 Hsu J, Santesso N, Mustafa R *et al.* Antivirals for treatment of influenza: a systematic review and meta-analysis of observational studies. *Ann Intern Med* 2012; 156:512–524.
- 12 Hernan MA, Lipsitch M. Oseltamivir and risk of lower respiratory tract complications in patients with flu symptoms: a meta-analysis of eleven randomized clinical trials. *Clin Infect Dis* 2011; 53:277–279.
- 13 Kumar D, Michaels MG, Morris MI *et al.* American Society of Transplantation H1N1 Collaborative Study Group. Outcomes from pandemic influenza A H1N1 infection in recipients of solid-organ transplants: a multicentre cohort study. *Lancet Infect Dis* 2010; 10:521–526.
- 14 Rodriguez A, Diaz E, Martin-Loeches I *et al.* Impact of early oseltamivir treatment on outcome in critically ill patients with 2009 pandemic influenza A. *J Antimicrob Chemother* 2011; 66:1140–1149.
- 15 Farias JA, Fernandez A, Monteverde E *et al.* Critically ill infants and children with influenza A (H1N1) in pediatric intensive care units in Argentina. *Intensive Care Med* 2010; 36:1015–1022.
- 16 Hiba V, Chowders M, Levi-Vinograd I, Rubinovitch B, Leibovici L, Paul M. Benefit of early treatment with oseltamivir in hospitalized patients with documented 2009 influenza A (H1N1): retrospective cohort study. *J Antimicrob Chemother* 2011; 66:1150–1155.
- 17 <http://www.thefederalregister.com/d.p/2009-10-02-E9-23761>
- 18 Darwish I, Mubareka S, Liles W. Immunomodulatory therapy for severe influenza. *Expert Rev Anti Infect Ther* 2011; 9:807–822.
- 19 Shaw M. The host interactome of influenza virus presents new potential targets for antiviral drugs. *Rev Med Virol* 2011; 21:358–369.
- 20 Beigel J, Harman LA, Collins PS *et al.* Pharmacokinetic and safety evaluations of escalating doses of peramivir administered intravenously in health volunteers. *ICAAC* 2007; Poster A-1408.
- 21 Furuta Y, Takakashi K, Kuno-Macakawa M *et al.* Mechanism of action of T-705 against influenza virus. *Antimicrob Agents Chemother* 2005; 49:981–986.
- 22 Ishizuka H, Yoshida S, Okabe H, Yoshihara K. Clinical pharmacokinetics of laninamivir, a novel long-acting neuraminidase inhibitor, after single and multiple inhaled doses of its prodrug, CS-8958, in healthy male volunteers. *J Clin Pharmacol* 2010; 51:1319–1329.
- 23 Iversen P, Vourich D, Schnell F, Thomas V. AVI-7100: Novel therapeutic in response to pandemic influenza (H1N1-SOIV). *ICAAC* 2010.
- 24 Ekiert D, Bhabha G, Elsliger M-A *et al.* Antibody recognition of a highly conserved influenza virus epitope. *Science* 2009; 324:246–251.
- 25 <http://www.autoimmune.com/influenzagen.html>
- 26 <http://www.evolva.com/products/pharmaceuticals/others>
- 27 Belardo G, La Frazia S, Cenciarelli O *et al.* Nitazoxanide, A Novel Potential Anti-Influenza Drug Acting in Synergism with Neuraminidase Inhibitors. 49th Annual Meeting of the Infectious Diseases Society of America. Boston, MA: Infectious Diseases Society of America, 2011.
- 28 Fry A. Oseltamivir-Resistant 2009 Pandemic Influenza A(H1N1) Virus (H1N1pdm09) Infections—United States, 2010–2011. Chicago: International Conference on Antimicrobial Agents and Chemotherapy, 2011.
- 29 Oseltamivir-resistant influenza A(H1N1)2009 cluster in Australia. Rapid Risk Assessment, European Centre for Disease Prevention and Control, 2011.
- 30 Ison M, de Jong M, Gilligan K *et al.* End points for testing influenza antiviral treatments for patients at high risk of severe and life-threatening disease. *J Infect Dis* 2010; 201:1654–1662.
- 31 <http://investor.shareholder.com/biocryst/releasedetail.cfm?ReleaseID=552784>
- 32 http://www.biota.com.au/uploaded/154/1021743_36bardaawards-majorcontrac.pdf