U.S. Department of Health and Human Services
Food and Drug Administration
Center for Drug Evaluation and Research
Office of Translational Sciences
Office of Biostatistics

STATISTICAL REVIEW AND EVALUATION

CLINICAL STUDIES

NDA Number:	22368
Drug Name:	Aridol (also known as Mannitol)
Indication(s):	"For assessment of bronchial hyperresponsiveness to aid in the diagnosis of patients ≥ 6 years of age with symptoms of or suggestive of asthma"
Applicant:	Pharmaxis
Date(s):	Stamp date: February 27, 2009
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Keywords:

NDA, clinical studies, diagnostic test

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1. EXECUTIVE SUMMARY

1.1 Conclusions and Recommendations

Study 305 adequately demonstrates that aridol provides better diagnostic ability than chance alone, that is, the sensitivity and specificity of the aridol test exceed 50%. The confidence intervals for the differences between aridol and methacholine in sensitivity and specificity illustrate the degree to which the two diagnostic tests are the same and can be used to make a judgment regarding whether the two test are providing noninferior levels of information. While the estimates of the differences in sensitivity and specificity between aridol and methacholine are numerically small, it is important to consider these differences in context of the small benefit over chance alone.

1.2 Brief Overview of Clinical Studies

The sponsor has conducted two (one non-US and one US) phase 3 clinical studies to support the regulatory approval of Aridol with the following proposed indication: "Aridol is an indirect bronchial challenge test indicated for the assessment of bronchial hyperresponsiveness to aid in the diagnosis of patients ≥ 6 years of age with symptoms of or suggestive of asthma". The sponsor indicates that the diagnostic effectiveness claim has been assessed primarily on the basis of the US study, study number DPM-A-305 (hereafter referred to within this document as study 305) and is supported by the non-US study, study number DPM-A-301 (hereafter referred to within this document as study 301).

The pivotal study, study 305, is titled, "A Phase III Multicenter Study to Demonstrate the Sensitivity and Specificity of Aridol (Mannitol) Challenge as Compared with Methacholine Challenge to Predict Bronchial Hyperresponsiveness as Manifested by a Positive Exercise Challenge in Subjects Presenting with Signs and Symptoms Suggestive of Asthma but Without a Definitive Diagnosis". As part of this study, subjects were independently diagnosed as positive or negative using the methacholine challenge test (with methacholine positivity defined as the dose of provoking stimulus causing a 20% fall in FEV_1 was less than or equal to either 12 mg/mL (SAP defined) or 16 mg/mL (ATS guidelines)) and using the aridol challenge test (with aridol positivity defined as the dose of provoking stimulus causing a 15% fall in FEV_1 was achieved at any dose until the maximum dose had been given or a between-dose drop of ≥ 10% in FEV_1 was observed). Subjects were also required to undergo two exercise challenge tests for diagnosis of exercise-induced bronchospasm (with exercise positivity defined as ≥10% fall in FEV_1 after either of two standardized treadmill runs) to act as the standard of truth for calculation of the sensitivity and specificity of the methacholine and aridol diagnostic tests. The primary objectives of the study were

(1.) to accurately estimate sensitivity and specificity of aridol challenge to detect bronchial hyperresponsiveness (BHR), i.e., within a 10% margin of the point estimates,
(2.) to demonstrate that aridol challenge sensitivity for BHR is significantly greater than 60%,
(3.) to demonstrate aridol challenge specificity is significantly greater than that seen with methacholine challenge to detect BHR (as manifested by a positive exercise challenge).

The supportive study, study 301, is titled, "A Phase 3 Study to Determine the Safety and Efficacy of Inhaled Dry Powder Manitol as a Bronchial Provocation Test for Airway Hyperresponsiveness". As part of this study, known asthmatic/symptomatic and known non-asthmatic subjects were enrolled and independently diagnosed as positive or negative using the aridol challenge test (with aridol positivity defined as the dose of provoking stimulus causing a 15% (or 10%) fall in FEV_1 was achieved at any dose until the maximum dose had been given) and by the respiratory physician using the hypertonic saline comparator challenge as well as the subject's respiratory and medical history (excluding the results of the aridol test). As part of the inclusion and exclusion criteria, asthmatic/symptomatic subjects were required to have active signs and symptoms of asthma (as defined by Asthma Management Handbook 2002 pg 4) and non-asthmatic subjects were required to have never had a clinical diagnosis of asthma nor experienced signs and symptoms suggestive of asthma. The primary efficacy objective of the study was to describe the sensitivity and specificity of aridol relative to the standard of truth, the respiratory physician diagnosis (which was based on the saline challenge as a bronchial provocation test and the respiratory and medical history). There was no comparator diagnostic procedure involved in this study.

1.3 Statistical Issues and Findings

The following statistical issues and their impact have been described in the context of the review. Please refer to the specified section for details.

Study 301

- As communicated to the sponsor at the pre-IND meeting held July 19, 2004, use of study 301 for regulatory purposes is significantly limited by flaws in the study design including an inappropriate target population and a lack of a comparator diagnostic procedure. (Section 2.1)

Study 305

- The target population for study 305 was subjects with an equivocal diagnosis of asthma or who had been referred for further investigation of asthma-type symptoms, a population that is likely more similar to the population for which the product is intended to be approved. In addition, study 305 is designed with a comparator procedure, methacholine challenge, a product that is FDA-approved for the indication sought for aridol. Therefore study 305 is considered more appropriate than study 301 for demonstration of the efficacy of aridol for regulatory purposes. (Sections 2.1 and 3.1.1)
- The protocol originally defined the ITT group as simply all subjects who received at least one dose of methacholine or aridol. This definition was not directly implemented by the sponsor; however, the "ITT plus" group seems to most closely represent this definition thus in this document, analysis of the ITT plus group, rather than the ITT group, is presented. Results of the efficacy analyses in the ITT group are largely consistent with those of the ITT plus group. (Section 3.1.2)
- Reviewer analyses and sponsor analyses numerically differ by a very small amount. The sponsor presents results using the Markov Chain Monte Carlo (MCMC) simulation citing that as per the statistical analysis plan, the outcome of the MCMC analyses were provided (in lieu of the more traditional use of the normal approximation methods) because the distributions for sensitivity and specificity were not absolutely normal.

However, with the large size of this study, it is unclear how the distributions of sensitivities and specificities could be dramatically non-normal. Therefore, this review provides results of the more traditional normal approximation methods, presenting, among other results, the 95% confidence intervals for the differences in sensitivities and specificities calculated based on McNemar's test for paired data. Given that the normal approximation methods were the primary methods specified for the efficacy analysis in the protocol and statistical analysis plan and that the distributions of the sensitivities and specificities in a large study should be adequately normal, the normal approximation methods are preferred and are presented in Table 1. Qualitative conclusions resulting from the two approaches are largely the same. (Section 3.1.2)

- Analyses addressing the primary efficacy objectives are presented under varying conditions. Since it was unclear in the protocol whether the ITT plus or PP group was considered primary, analyses in both groups are presented. Analyses implementing varying cutoffs for the methacholine challenge (16 mg/mL, the standard published in the ATS guidelines, 12 mg/mL, as specified in the statistical analysis plan, and 4 mg/mL due to interest from the FDA medical review team) are presented. In general, a worst case approach was used for addressing missing data; however, an analysis ignoring the missing data is also presented. In general, none of these criteria dramatically impacted the results of the analyses. (Sections 3.1.1 and 3.1.2)
- Although two of the three a priori documented study objectives were not met, the efficacy of aridol for regulatory purposes may still be substantiated by this study in that, it may be argued that the primary efficacy objectives defined as part of this study are not the most relevant in terms of assessing the efficacy of the aridol challenge test for regulatory approval. (Section 3.1.2)
- Prior to NDA submission, the Division expressed an interest in demonstrating that the sensitivity and specificity of aridol challenge are similar to that of a comparator procedure. This requires definition of the clinical meaning of *similarity* in sensitivity and specificity. As this study was not designed with this noninferiority objective in mind, no a priori noninferiority margin was documented in the protocol. In the absence of such documentation, we use the confidence intervals for the differences between aridol and methacholine in sensitivity and specificity to simply illustrate the degree to which the two diagnostic tests are the same and leave to clinical judgment whether this level of precision is acceptable in order to conclude that the two procedures are providing analogous levels of information. (Section 3.1.2)
- At the request of the FDA medical review team, plots of the cumulative dose of aridol or methacholine by the mean percent change from baseline in FEV_1 for the exercise positive and exercise negative strata are provided in this review. However, the use of these plots may be limited since the *mean* percent change from baseline in FEV_1 may be a misleading endpoint in this setting. First, the mean result may not be a good indicator for what will happen to a typical individual subject. And second, since subjects with the greatest falls in FEV_1 at the lower cumulative doses do not proceed to the higher cumulative doses (as they are diagnosed as positive and dosing stops), the impact of missing data on the mean fall in FEV_1 becomes more pronounced for the higher cumulative doses. In lieu of examining the *mean* percent change from baseline in FEV_1, plots of the percent change from baseline in FEV_1 for each individual could be considered. Such plots are provided in Appendix II. (Section 3.1.2 and Appendix II)

- The primary efficacy analyses by age, gender, and race are provided. No differing treatment effects among the subgroups examined were noted. (Section 4.1 and Appendix I)

2. INTRODUCTION

2.1 Overview

The sponsor has conducted two (one non-US and one US) phase 3 clinical studies to support the regulatory approval of Aridol with the following proposed indication: "Aridol is an indirect bronchial challenge test indicated for the assessment of bronchial hyperresponsiveness to aid in the diagnosis of patients ≥ 6 years of age with symptoms of or suggestive of asthma". The sponsor indicates that the diagnostic effectiveness claim has been assessed primarily on the basis of the US study, study number DPM-A-305 (hereafter referred to within this document as study 305) and is supported by the non-US study, study number DPM-A-301 (hereafter referred to within this document as study 301).

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The supportive study, study 301, is titled, "A Phase 3 Study to Determine the Safety and Efficacy of Inhaled Dry Powder Manitol as a Bronchial Provocation Test for Airway Hyperresponsiveness". As part of this study, known asthmatic/symptomatic and known non-asthmatic subjects were enrolled and independently diagnosed as positive or negative using the aridol challenge test (with aridol positivity defined as the dose of provoking stimulus causing a 15% (or 10%) fall in FEV_1 was achieved at any dose until the maximum dose had been given) and by the respiratory physician using the hypertonic saline comparator challenge as well as the subject's respiratory and medical history (excluding the results of the aridol test). As part of the

inclusion and exclusion criteria, asthmatic/symptomatic subjects were required to have active signs and symptoms of asthma (as defined by Asthma Management Handbook 2002 pg 4) and non-asthmatic subjects were required to have never had a clinical diagnosis of asthma nor experienced signs and symptoms suggestive of asthma. The primary efficacy objective of the study was to describe the sensitivity and specificity of aridol relative to the standard of truth, the respiratory physician diagnosis (which was based on the saline challenge as a bronchial provocation test and the respiratory and medical history). There was no comparator diagnostic procedure involved in this study.

Communication with the sponsor regarding these studies is documented under pre-IND 70277. Pertinent parts of the statistical portion of those communications are summarized herein.

Study 301 was discussed with the sponsor at a pre-IND meeting held July 19, 2004. The sponsor provided the following question "Is the proposed single phase 3 study and its design adequate to support an NDA filing". The Division responded that "A single pivotal study may be adequate to support filing an NDA. However, the proposed phase 3 protocol is not adequately designed to meet its objectives". In addition to noting the clinical concern that hypertonic saline is not the gold standard for the detection of bronchial hyperresponsiveness, the Division described the statistical deficiencies in the protocol as follows

(1.) The protocol proposes to enroll a group of "known asthmatics" and a group of non-asthmatic subjects required to "have never had a clinical diagnosis of asthma nor experienced signs and symptoms suggestive of asthma". The sensitivity and specificity of the mannitol provocation test in a group of subjects with a known diagnosis of asthma may not be indicative of the performance of the test in a group of subjects with suspected asthma but whose diagnosis is not established. Since the later is the group likely to receive the diagnostic test, examination of the sensitivity and specificity in that type of a patient group is necessary to support approval.

(2.) As designed, the study will provide point estimates of the sensitivity and specificity (and their 95% confidence intervals) of the mannitol provocation test; however, because these estimates can be affected by the spectrum of the study subjects' disease, it will be necessary to consider these performance measures relative to those of another diagnostic procedure, such a methacholine challenge. Therefore, an appropriate study design for evaluation of a diagnostic test should include a statistical comparison of the sensitivities and specificities of each of the diagnostic procedures (mannitol challenge and methacholine challenge, for example) where the sensitivity and specificity of each challenge can be calculated relative to some gold standard, perhaps a standardized assessment of clinical diagnosis of asthma.

(3.) The proposed study protocol indicates that the primary efficacy analysis will include the subset of randomized subjects who satisfy all inclusion and exclusion criteria and complete both challenges. Since exclusion of subjects based on post-randomization findings may yield a biased subset, we also consider the efficacy results of the intent-to-treat (ITT) group, which includes all patients who were randomized. For subjects in the ITT group with missing efficacy data, a worst-case approach would be used for imputation of their results. If the diagnosis according to the mannitol (or comparator) challenge is not available for this analysis, that subject should be considered to have been incorrectly diagnosed by the mannitol (or comparator) test.

The sponsor subsequently indicated that after the Agency informed them at the July 19, 2004 pre-IND meeting that study 301 was not sufficient to establish the efficacy of aridol for the proposed indication, a second phase 3 study, study 305, was conducted following the Agency's above recommendations.

Study 305, the study designated as pivotal by the sponsor, will be thoroughly reviewed within this document. However, given the significant limitations of study 301 described above (i.e., inappropriate target population and lack of a comparator diagnostic procedure) the review of study 301 (a study which is described by the sponsor as a supportive study) will be limited to discussion of the inadequacies in the design.

2.2 Data Sources

The following data sets were requested by the Division, submitted electronically by the sponsor, and utilized in the review of this study.

//cdsesub1/evsprod/NDA022368//0004/m5/datasets/dpm-a-305/tabulations/request1.xpt
//cdsesub1/evsprod/NDA022368//0004/m5/datasets/dpm-a-305/tabulations/request2.xpt

All submitted data sets were found to be adequately documented and organized.

3. STATISTICAL EVALUATION

3.1 Evaluation of Efficacy

3.1.1 Study Design (Study 305)

The pivotal study, study 305, is titled, "A Phase III Multicenter Study to Demonstrate the Sensitivity and Specificity of Aridol (Mannitol) Challenge as Compared with Methacholine Challenge to Predict Bronchial Hyperresponsiveness as Manifested by a Positive Exercise Challenge in Subjects Presenting with Signs and Symptoms Suggestive of Asthma but Without a Definitive Diagnosis". As part of this study, subjects were independently diagnosed as positive or negative using the methacholine challenge test (with methacholine positivity defined as the dose of provoking stimulus causing a 20% fall in FEV_1 was less than or equal to either 12 mg/mL (SAP defined) or 16 mg/mL (ATS guidelines)) and using the aridol challenge test (with aridol positivity defined as the dose of provoking stimulus causing a 15% fall in FEV_1 was achieved at any dose until the maximum dose had been given or a between-dose drop of $\geq$ 10% in FEV_1 was observed). Subjects were also required to undergo two exercise challenge tests for diagnosis of exercise-induced bronchospasm (with exercise positivity defined as $\geq$10% fall in FEV_1 after either of two standardized treadmill runs) to act as the standard of truth for calculation of the sensitivity and specificity of the methacholine and aridol diagnostic tests. The primary objectives of the study were
(1.) to accurately estimate sensitivity and specificity of aridol challenge to detect bronchial hyperresponsiveness (BHR), i.e., within a 10% margin of the point estimates,
(2.) to demonstrate that aridol challenge sensitivity for BHR is significantly greater than 60%,

(3.) to demonstrate aridol challenge specificity is significantly greater than that seen with methacholine challenge to detect BHR (as manifested by a positive exercise challenge).

The target population for this study was subjects with an equivocal diagnosis of asthma or who had been referred for further investigation of asthma-type symptoms. Subjects were required to be between 6 and 50 years of age. Subjects with chronic restrictive or obstructive pulmonary diseases (cystic fibrosis, COPD, bronchiectasis, chronic bronchitis, emphysema, tuberculosis, pulmonary carcinoma, pulmonary fibrosis, pulmonary hypertension, hypercapnia) were excluded from the study. In total, the protocol specified eight inclusion and 24 exclusion criteria for enrollment in this study. Inclusion and exclusion criteria were to be assessed at visit 1, the screening visit.

Subjects were to undergo an exercise challenge test for diagnosis of exercise induced bronchospasm at both visits 2 and 3. Visit 2 was to occur 1 to 4 days after the screening visit and visit 3 was to occur 1 to 4 days after visit 2, each at a recommended starting time within ±2 hours of the starting time of the screening visit. Subjects with a positive outcome for at least one of two exercise challenge tests were considered "exercise positive" for purposes of the standard of truth for this study and were therefore to be used in the calculation of the sensitivities of interest. Subjects with a negative outcome on both exercise challenge tests were considered "exercise negative" for purposes of the standard of truth for this study and were therefore to be used in the calculation of specificities of interest.

The aridol challenge and methacholine challenge were each to be administered at visit 4 or 5. Visit 4 was to occur 1 to 4 days after the visit 3 and visit 5 was to occur 1 to 4 days after visit 4, each at a recommended starting time within ±2 hours of the starting time of the screening visit. Randomizations of the order of administration of the aridol and methacholine challenge test were 1:1 and were completed separately for the exercise positive and exercise negative groups. To maintain blinding, the aridol and methacholine challenges were performed by personnel separate from the screening assessment team and respiratory physician. The results of the challenge tests were not disclosed to the assessment team or the respiratory physician.

- Aridol Challenge: The aridol challenge was administered as a diagnostic test for BHR. The total dose administered ranged from 0 mg to 635 mg, depending on airway response. Aridol was given sequentially as follows: 0 mg, 5 mg, 10 mg, 20 mg, 40 mg, 80 mg, 160 mg, 160 mg, and 160 mg. Each dose followed the previous dose until the FEV_1 fell by ≥ 15% from baseline, a between-dose fall in FEV_1 was ≥ 10%, or the cumulative dose of 635 mg had been administered. The provoking dose of aridol to induce the 15% fall in FEV_1 (i.e., PD_{15}) was calculated by linear interpolation from the curve relating the percent fall in FEV_1 from the post 0 mg capsule baseline value for FEV_1 to the cumulative dose of aridol delivered (e.g., 5 mg, 15, mg, 35 mg, 75 mg, 155 mg, 315 mg, 475 mg, or 635 mg). For purposes of this study, aridol positivity was defined as the PD_{15} being achieved by the maximum dose or a between-dose drop of ≥ 10% in FEV_1 was observed.
- Methacholine Challenge: The methacholine challenge was administered as a diagnostic test for BHR. Methacholine was given sequentially as follows: 0.0312 mgh/mL, 0.0625 mg/mL, 0.125 mg/mL, 0.25 mg/mL, 0.5 mg/mL, 1 mg/mL, 2 mg.mL, 4 mg/mL, 8 mg/mL, 16 mg/mL. Each dose followed the previous dose

until the FEV_1 fell by ≥ 20% from baseline or until all doses had been administered. The provoking dose of methacholine to induce the 20% fall in FEV_1 (i.e., PD_{20}) was calculated by linear interpolation from the curve relating the percent fall in FEV_1 from the baseline value for FEV_1 to the cumulative dose of methacholine delivered. For purposes of this study, methacholine positivity was defined as the PD_{20} being less than or equal to either 12 mg/mL (SAP defined) or 16 mg/mL (ATS guidelines)).

Also at visit 5, a respiratory physician, a clinician, diagnosed the subjects (by examining the subject and reviewing the subject's study record including any relevant diagnostic information available at the time of this visit except the methacholine or aridol challenge tests). Subjects were classified into one of the following categories

- asthma is extremely likely or definite (95 to 100% likelihood)
- asthma is very likely (72.5 to <95% likelihood)
- asthma is probable (50 to 72.5% likelihood)
- asthma is possible (27.5 to <50% likelihood)
- asthma is unlikely but cannot be excluded (5 to <27.5% likelihood)
- asthma is very unlikely or excluded (0 to <5% likelihood)

The intent-to-treat (ITT) group was protocol-defined as all subjects who received at least one dose of methacholine or aridol. The per-protocol population was protocol-defined as all subjects with no major protocol violations that complete all of the required challenge tests, including methacholine and aridol challenges. The primary efficacy analysis was to be conducted in both the ITT and PP groups. It was not clear from the protocol which analysis group was to be considered primary. Missing diagnoses for the aridol or methacholine challenges were to be imputed using a worst-case approach as follows: missing aridol diagnoses were assumed to be negative if the subject was exercise positive and positive if the subject was exercise negative while missing methacholine diagnoses were assumed to be positive if the subject was exercise positive and negative if the subject was exercise negative.

The primary efficacy analysis specified in the statistical analysis plan was to calculate 95% confidence intervals for the sensitivities and specificities of the aridol and methacholine challenges using normal approximations for the binomial distribution. In addition, 95% confidence intervals for the differences between aridol and methacholine in sensitivity and specificity were to be calculated using normal-approximations for the binomial distribution. Each of the three efficacy objectives stated above for this study would then have been considered successfully achieved if the following criteria were met:

(1.) both the lower and upper confidence interval limits for the sensitivity and specificity of aridol challenge should be within a 10% points of the point estimates,

(2.) the lower confidence interval limit for the aridol challenge sensitivity should be greater than 60%,

(3.) the lower confidence interval limit for the difference in aridol challenge specificity and the methacholine challenge specificity is greater than zero.

The statistical analysis plan noted that if the bivariate distributions of sensitivities and specificities are visibly non-normal, then tests for the primary objectives would be conducted

through Markov Chain Monte Carlo (MCMC) simulation. No correction for multiplicity was planned for the three primary objectives as success with all three was required for successful demonstration of the efficacy of the aridol challenge.

3.1.2 Results (Study 305)

Five hundred nine unique subjects were screened for enrollment in study 305. Seventy three were not enrolled due to events occurring prior to randomization (including inclusion/exclusion criteria not met (49), withdrew consent/lost to follow-up (15), excess FEV_1 variability (1), adverse event (1), and enrollment closed (7)) leaving 436 in the all-randomized / safety analysis group. An additional 16 subjects were excluded from the efficacy analyses post-randomization (withdrew consent (5), took prohibited drug (2), excess FEV_1 variability (1), adverse event (2), and enrollment closed (2)) leaving 420 (96%) in the "intent-to-treat plus" (ITT plus) group. The ITT plus group included 29 subjects whose exercise challenges were both negative but were considered inadequate. Therefore the sponsor also defined an ITT group excluding these 29 subjects from the 420 in the ITT plus group therefore leaving 391 (90%) subjects. An additional 16 subjects are excluded from the ITT group to create the per protocol (PP) group with 375 (86%) subjects. Finally, a "PP plus" group was created by adding the 29 subjects with exercise challenges that were negative but considered inadequate to the 375 PP subjects therefore including 404 (93%) subjects.

The protocol originally defined the ITT group as simply all subjects who received at least one dose of methacholine or aridol. The ITT plus group (i.e., including the 29 subjects with negative but possibly inadequate exercise challenges) seems to most closely represent this definition thus in this document, analysis of the ITT plus group, rather than the ITT group, is presented. However, examination of the efficacy analyses in the ITT group was also undertaken as part of this review and the conclusions from those analyses are largely consistent with those of the ITT plus group. In addition, this review presents analyses of the PP group being that this group most closely represents the protocol definition for the PP group which was all subjects with no major protocol violations that complete all of the required challenge tests, including methacholine and aridol challenges. As with the ITT plus and ITT efficacy analyses, the PP plus and PP efficacy analyses yield largely consistent conclusions.

The analyses necessary to address the primary efficacy objectives for this study are included in Table 1 (and in more detail in appendix 1). These include the sensitivities and specificities (calculated relative to exercise challenge) and the associated 95% confidence intervals for aridol and methacholine as well as the differences in these measures between aridol and methacholine and the associated 95% confidence intervals.

The results in Table 1 are reviewer analyses and numerically differ by a very small amount from the results presented by the sponsor in the study report. The sponsor presents results using the Markov Chain Monte Carlo (MCMC) simulation citing that as per the statistical analysis plan, the outcome of the MCMC analyses were provided (in lieu of the more traditional use of the normal approximation methods) because the distributions for sensitivity and specificity were not absolutely normal. However, with the large size of this

study, it is unclear how the distributions of sensitivities and specificities could be dramatically non-normal. Therefore, this review provides results of the more traditional normal approximation methods, presenting, among other results, the 95% confidence intervals for the differences in sensitivities and specificities calculated based on McNemar's test for paired data. Given that the normal approximation methods were the primary methods specified for the efficacy analysis in the protocol and statistical analysis plan and that the distributions of the sensitivities and specificities in a large study should be adequately normal, the normal approximation methods are preferred and are presented in Table 1. Qualitative conclusions resulting from the two approaches are largely the same.

Table 1: By-Treatment Group Comparisons of Sensitivity & Specificity (Calculated Relative to Exercise Challenge) for Aridol and Methacholine for Assessment of the Primary Efficacy Objectives

Conditions			Sensitivity (95% CI)			Specificity (95% CI)		
Analysis Group	Methacholine Cutoff	Missing Data	Aridol	Methacholine	Difference	Aridol	Methacholine	Difference
ITT plus	16	Worst Case	58% (50%, 65%)	53% (46%, 61%)	5% (-4%, 13%)	63% (57%, 69%)	68% (62%, 73%)	-5% (-12%, 3%)
ITT plus	16	Ignored	58% (51%, 66%)	54% (46%, 61%)	5% (-4%, 13%)	64% (58%, 70%)	68% (62%, 73%)	-4% (-11%, 3%)
ITT plus	12	Worst Case	58% (50%, 65%)	50% (43%, 58%)	7% (-2%, 16%)	63% (57%, 69%)	72% (67%, 78%)	-9% (-16%, -2%)
ITT plus	4	Worst Case	58% (50%, 65%)	35% (28%, 43%)	22% (14%, 31%)	63% (57%, 69%)	84% (79%, 89%)	-21% (-27%, -14%)
PP	16	Worst Case	58% (51%, 66%)	55% (48%, 63%)	3% (-6%, 12%)	65% (58%, 71%)	69% (63%, 75%)	-4% (-12%, 3%)
PP	12	Worst Case	58% (51%, 66%)	52% (44%, 60%)	6% (-3%, 15%)	65% (58%, 71%)	74% (68%, 80%)	-9% (-16%, -2%)

Source: reviewer analyses

The analyses in Table 1 are presented under varying conditions. Since it was unclear in the protocol whether the ITT plus or PP group was considered primary, analyses in both groups are presented. Analyses implementing cutoffs for the methacholine challenge of 16 mg/mL, the standard published in the ATS guidelines, 12 mg/mL, as specified in the statistical analysis plan, and 4 mg/mL due to interest from the FDA medical review team are presented. In general, a worst case approach was used for addressing missing data as follows: missing aridol diagnoses were assumed to be negative if the subject was exercise positive and positive if the subject was exercise negative while missing methacholine diagnoses were assumed to be positive if the subject was exercise positive and negative if the subject was exercise negative. Given the very conservative nature of this missing data imputation, an analysis ignoring the missing data is also presented to illustrate whether the missing data imputation was severely affecting the overall conclusions of the analyses. With the exception of the case where a methacholine cutoff of 4 mg/mL was used, as is shown in Table 1, none of these criteria dramatically altered the results of the analyses.

The sponsor's primary objectives for this study were

(1.) to accurately estimate sensitivity and specificity of aridol challenge to detect bronchial hyperresponsiveness (BHR), i.e., within a 10% margin of the point estimates,

(2.) to demonstrate that aridol challenge sensitivity for BHR is significantly greater than 60%,

(3.) to demonstrate aridol challenge specificity is significantly greater than that seen with methacholine challenge to detect BHR (as manifested by a positive exercise challenge).

As illustrated by both the lower and upper confidence interval limits for the sensitivity and specificity of aridol challenge being within 10 percentage points of the point estimates, the analyses in Table 1 confirm the sponsor's first study objective in all cases presented. The sponsor's second study objective is not confirmed for any case presented in Table 1 as illustrated by the lower confidence interval limit for the aridol challenge sensitivity being less than 60%. And finally, the sponsor's third study objective is also not confirmed for any case presented in Table 1 as illustrated by the lower confidence interval limit for the difference in aridol challenge specificity and the methacholine challenge specificity being less than zero. Thus on their face, it appears that the primary efficacy analyses present in Table 1 may not support the efficacy of an aridol challenge test.

However, it may be argued that the primary efficacy objectives defined as part of this study are not the most relevant in terms of assessing the efficacy of the aridol challenge test for regulatory approval. At a pre-IND meeting held July 19, 2004, the sponsor was advised that the Division believed that an appropriate study design for evaluation of a diagnostic test should include a statistical comparison of the sensitivity and specificity of the new diagnostic procedure with an established/FDA-approved diagnostic procedure (methacholine challenge, for example) where the sensitivity and specificity of each challenge are calculated relative to some gold standard. The goal of such an approach would be two-fold. First, the new diagnostic procedure should perform better than chance alone, that is, the sensitivity and specificity and the new diagnostic procedure should exceed 50%. And second, the new diagnostic procedure must be shown to possess sensitivity and specificity *similar* to that of the FDA-approved procedure. Although these objectives were not adopted a priori, the study design for study 305 allows such comparisons.

Again referring to Table 1, the lower confidence interval limits for the sensitivities and specificities for aridol being (marginally) greater than 50% illustrate that the first of these post-hoc objectives is achieved. The second post-hoc objective is more difficult to asses because it requires definition of the meaning of *similarity* in sensitivity and specificity. As this study was not designed with this noninferiority objective in mind, no a priori noninferiority margin was documented in the protocol. In the absence of such documentation, we use the data in Table 1, specifically the confidence intervals for the differences between aridol and methacholine in sensitivity and specificity to simply illustrate the degree to which the two diagnostic tests are the same and leave to clinical judgment whether this level of precision is acceptable in order to conclude that the two procedures are providing analogous levels of information. Taking the first case from Table 1 (i.e., ITT plus, methacholine cutoff of 16 mg/mL, and worst case missing data imputation) as an example, the sensitivity of aridol is demonstrated to be no more than 4 percentage points worse (and may be up to 13 percentage points better) than the sensitivity of methacholine while the specificity of aridol is demonstrated to be no more than 12 percentage points worse (and may be up to 3 percentage points better) than the specificity of methacholine. While these differences are numerically small, it is important to consider these differences in the context of the small benefit over chance alone. With the exception of the case where a methacholine cutoff of 4 mg/mL was used, the results of the other cases are generally comparable to this.

Table 2 (and in more detail appendix 1) presents analyses that are analogous to those presented in Table 1 with the exception that the blinded physician diagnosis made at visit 5 is used as the standard of truth rather than the results of the exercise challenges. Physician diagnosis of "probably", "possible", "very likely", and "extremely likely or definite" were considered positive diagnoses and "unlikely but not excluded" and "very unlikely or excluded" were considered negative diagnoses for purposes of this analyses. These secondary analyses are being presented due to interest generated by the FDA-review team. These analyses can be interpreted in the same way as described above for the analyses provided in Table 1.

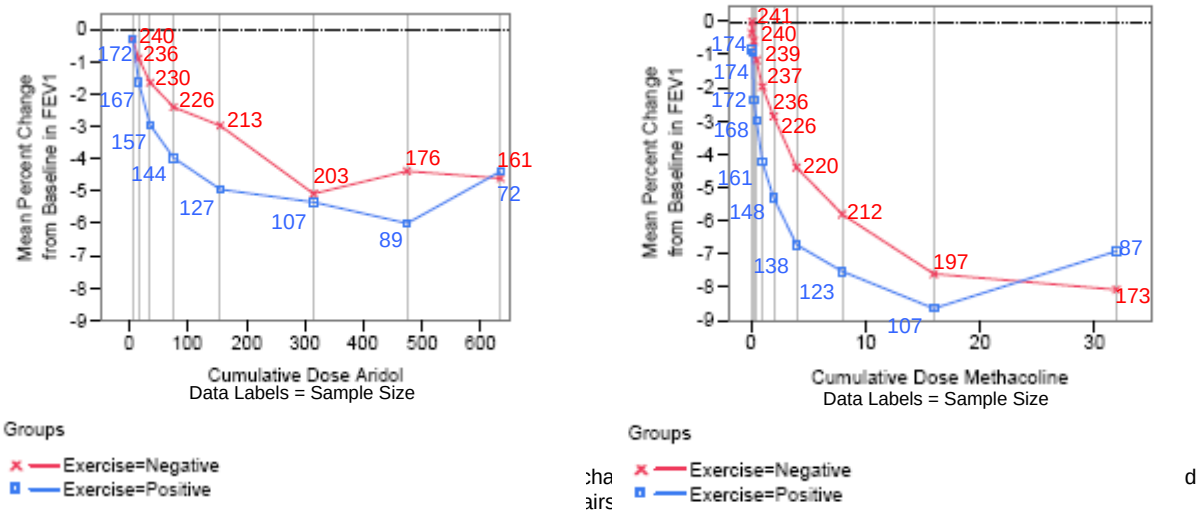
Table 2: By-Treatment Group Comparisons of Sensitivity & Specificity (Calculated Relative to Blinded Physician Diagnosis from Visit 5) for Aridol and Methacholine

Conditions			Sensitivity (95% CI)			Specificity (95% CI)		
Analysis Group	Methacholine Cutoff	Missing Data	Aridol	Methacholine	Difference	Aridol	Methacholine	Difference
ITT plus	16	Worst Case	54% (48%, 60%)	50% (44%, 56%)	4% (-3%, 11%)	69% (62%, 76%)	72% (65%, 79%)	-3% (-12%, 6%)
ITT plus	12	Worst Case	54% (48%, 60%)	45% (39%, 51%)	9% (2%, 16%)	69% (62%, 76%)	75% (68%, 81%)	-10% (-25%, 5%)
PP	16	Worst Case	55% (49%, 61%)	51% (45%, 57%)	4% (-3%, 11%)	73% (65%, 80%)	75% (67%, 82%)	-2% (-11%, 7%)
PP	12	Worst Case	55% (49%, 61%)	46% (40%, 52%)	9% (2%, 16%)	73% (65%, 80%)	77% (70%, 84%)	-4% (-13%, 4%)

Source: reviewer analyses

At the request of the FDA medical review team, plots of the cumulative dose of aridol or methacholine by the mean percent change from baseline in FEV_1 for the exercise positive and exercise negative strata are provided in Figure 1. The graphs are intended to illustrate that the fall in FEV_1 associated with administration of aridol is greater in the exercise positive subjects that in the exercise negative subjects and that this relationship is similar to that when methacholine is administered. While there is generally no statistically significant difference between the exercise positive and exercise negative groups in the mean percent change from baseline in FEV_1 for either aridol or methacholine, numerically, it does appear that the exercise positive subjects do experience a larger mean drop in FEV_1 than exercise negative subjects with administration of either product as evidenced by the blue lines generally falling below the red lines in Figure 1. From a statistical perspective though, the *mean* percent change from baseline in FEV_1 may be a misleading endpoint in this setting. First, the mean result may not be a good indicator for what will happen to a typical individual subject. And in considering a diagnostic test, it is necessary to diagnose an individual subject (i.e., dichotomization of the result for the individual subject, not the mean result across subjects, into positive or negative is needed). And second, since subjects with the greatest falls in FEV_1 at the lower cumulative doses do not proceed to the higher cumulative doses (as they are diagnosed as positive and dosing stops), the impact of missing data on the mean fall in FEV_1 becomes more pronounced for the higher cumulative doses. To allow fair comparisons between the exercise positive and exercise negative groups as well as between aridol and methacholine, it is necessary to assume that the impact of the missing data is equal in all these cases, a standard which is difficult to justify with the data available. In lieu of examining the *mean* percent change from baseline in FEV_1, plots of the percent change from baseline in FEV_1 for each individual could be considered. Such plots are provided in Appendix II.

Figure 1: Mean Percent Change from Baseline in FEV1 with Aridol or Methacholine by Exercise Stratum (ITT plus analysis group*)



3.1.3 Study Design and Discussion (Study 301)

Study 301 is titled, "A Phase 3 Study to Determine the Safety and Efficacy of Inhaled Dry Powder Manitol as a Bronchial Provocation Test for Airway Hyperresponsiveness". As part of this study, known asthmatic/symptomatic and known non-asthmatic subjects were enrolled and independently diagnosed as positive or negative using the aridol challenge test (with aridol positivity defined as the dose of provoking stimulus causing a 15% (or 10%) fall in FEV_1 was achieved at any dose until the maximum dose had been given) and by the respiratory physician using the hypertonic saline comparator challenge as well as the subject's respiratory and medical history (excluding the results of the aridol test). As part of the inclusion and exclusion criteria, asthmatic/symptomatic subjects were required to have active signs and symptoms of asthma (as defined by Asthma Management Handbook 2002 pg 4) and non-asthmatic subjects were required to have never had a clinical diagnosis of asthma nor experienced signs and symptoms suggestive of asthma. The primary efficacy objective of the study was to describe the sensitivity and specificity of aridol relative to the standard of truth, the respiratory physician diagnosis (which was based on the saline challenge as a bronchial provocation test and the respiratory and medical history). There was no comparator diagnostic procedure involved in this study.

As was discussed with the sponsor at a pre-IND meeting held July 19, 2004, there are significant statistical deficiencies in the design of study 301, most notably, first the study enrolled a patient population that was different from the population for which the product is intended to be approved and second the study did not have a comparator diagnostic procedure.

The study enrolled "known asthmatics" and a group of non-asthmatic subjects required to "have never had a clinical diagnosis of asthma nor experienced signs and symptoms suggestive of asthma". The sensitivity and specificity of aridol in a group of subjects with a known diagnosis may not be indicative of the performance of the test in a group of subjects with suspected asthma but whose diagnosis is not established. Since the later is the population in which regulatory approval is sought, examination of the sensitivity and specificity in that type of a patient group should be necessary to support approval.

Because the sensitivity and specificity estimates for aridol can be affected by the spectrum of the study subjects' disease, it is necessary to consider these performance measures for aridol relative to a comparator diagnostic procedure. The lack of a comparator in this study is akin to an uncontrolled clinical trial of a therapeutic agent with all subjects enrolled into a single group and receiving the experimental agent, allowing no comparison to placebo or an active treatment.

Due to these deficiencies in design, the efficacy data resulting from study 301 is, by design, not sufficient for regulatory approval. Therefore, this review focuses primarily on the more appropriately designed study for regulatory purposes, study 305.

4. FINDINGS IN SPECIAL/SUBGROUP POPULATIONS

4.1 Gender, Race and Age

The primary efficacy analyses by age, gender, and race for study 305 are given in Table 3 (and in more detail in appendix 1). No differing treatment effects among the subgroups examined were noted.

Table 3: By-Treatment Group Comparisons of Sensitivity & Specificity (Calculated Relative to Exercise Challenge) by Age, Gender, and Race

Ages 6 to 11 Years								
Conditions			Sensitivity (95% CI)			Specificity (95% CI)		
Analysis Group	Methacholine Cutoff	Missing Data	Aridol	Methacholine	Difference	Aridol	Methacholine	Difference
ITT plus	16	Worst Case	67% (47%, 87%)	71% (52%, 91%)	-5% (-29%,20%)	47% (21%, 72%)	33% (9%, 57%)	17% (-29%, 62%)
ITT plus	12	Worst Case	67% (47%, 87%)	67% (47%, 87%)	0% (-26%,26%)	47% (21%, 72%)	40% (15%, 65%)	7% (-32%, 46%)
Ages 12 to 17 Years								
Conditions			Sensitivity (95% CI)			Specificity (95% CI)		
Analysis Group	Methacholine Cutoff	Missing Data	Aridol	Methacholine	Difference	Aridol	Methacholine	Difference
ITT plus	16	Worst Case	55% (37%, 72%)	65% (48%, 81%)	-10% (-32%,13%)	62% (46%, 77%)	64% (49%, 79%)	-3% (-24%, 19%)
ITT plus	12	Worst Case	55% (37%, 72%)	65% (48%, 81%)	-10% (-32%,13%)	62% (46%, 77%)	74% (61%, 88%)	-13% (-32%, 6%)
Ages 17 Years and Above								
Conditions			Sensitivity (95% CI)			Specificity (95% CI)		
Analysis Group	Methacholine Cutoff	Missing Data	Aridol	Methacholine	Difference	Aridol	Methacholine	Difference
ITT plus	16	Worst Case	57% (48%, 65%)	47% (38%, 55%)	10% (1%, 20%)	65% (58%, 71%)	71% (65%, 77%)	-6% (-14%, 1%)
ITT plus	12	Worst Case	57% (48%, 65%)	44% (35%, 52%)	13% (3%, 23%)	65% (58%, 71%)	74% (68%, 80%)	-10% (-17%, -2%)
Males								
Conditions			Sensitivity (95% CI)			Specificity (95% CI)		
Analysis Group	Methacholine Cutoff	Missing Data	Aridol	Methacholine	Difference	Aridol	Methacholine	Difference
ITT plus	16	Worst Case	58% (47%, 68%)	47% (36%, 58%)	10% (-3%, 22%)	62% (54%, 71%)	69% (61%, 78%)	-7% (-17%, 3%)
ITT plus	12	Worst Case	58% (47%, 68%)	46% (35%, 57%)	11% (-2%, 24%)	62% (54%, 71%)	71% (63%, 79%)	-9% (-19%, 2%)
Females								
Conditions			Sensitivity (95% CI)			Specificity (95% CI)		
Analysis Group	Methacholine Cutoff	Missing Data	Aridol	Methacholine	Difference	Aridol	Methacholine	Difference
ITT plus	16	Worst Case	59% (49%, 68%)	59% (49%, 68%)	0% (-12%,12%)	64% (55%, 72%)	66% (58%, 74%)	-2% (-13%, 8%)
ITT plus	12	Worst Case	59% (49%, 68%)	54% (44%, 64%)	4% (-8%, 17%)	64% (55%, 72%)	73% (66%, 81%)	-9% (-19%, 0%)
Caucasian								
Conditions			Sensitivity (95% CI)			Specificity (95% CI)		
Analysis Group	Methacholine Cutoff	Missing Data	Aridol	Methacholine	Difference	Aridol	Methacholine	Difference
ITT plus	16	Worst Case	58% (50%, 67%)	57% (48%, 65%)	2% (-8%, 12%)	63% (56%, 70%)	67% (60%, 74%)	-4% (-12%, 4%)
ITT plus	12	Worst Case	58% (50%, 67%)	52% (43%, 61%)	6% (-4%, 17%)	63% (56%, 70%)	73% (66%, 79%)	-10% (-17%, -2%)
Non-Caucasian								
Conditions			Sensitivity (95% CI)			Specificity (95% CI)		
Analysis Group	Methacholine Cutoff	Missing Data	Aridol	Methacholine	Difference	Aridol	Methacholine	Difference
ITT plus	16	Worst Case	57% (42%, 71%)	46% (31%, 60%)	11% (-6%, 28%)	64% (51%, 76%)	69% (57%, 81%)	-5% (-21%, 11%)
ITT plus	12	Worst Case	57% (42%, 71%)	46% (31%, 60%)	11% (-6%, 28%)	64% (51%, 76%)	71% (59%, 82%)	-7% (-23%, 10%)

Source: reviewer analyses

4.2 Other Special/Subgroup Populations

No other special subgroups were identified for analysis in the course of this review.

5. SUMMARY AND CONCLUSIONS

5.1 Statistical Issues and Collective Evidence

The following statistical issues and their impact have been described in the context of the review. Please refer to the specified section for details.

Study 301

- As communicated to the sponsor at the pre-IND meeting held July 19, 2004, use of study 301 for regulatory purposes is significantly limited by flaws in the study design including an inappropriate target population and a lack of a comparator diagnostic procedure. (Section 2.1)

Study 305

- The target population for study 305 was subjects with an equivocal diagnosis of asthma or who had been referred for further investigation of asthma-type symptoms, a population that is likely more similar to the population for which the product is intended to be approved. In addition, study 305 is designed with a comparator procedure, methacholine challenge, a product that is FDA-approved for the indication sought for aridol. Therefore study 305 is considered more appropriate than study 301 for demonstration of the efficacy of aridol for regulatory purposes. (Sections 2.1 and 3.1.1)
- The protocol originally defined the ITT group as simply all subjects who received at least one dose of methacholine or aridol. This definition was not directly implemented by the sponsor; however, the "ITT plus" group seems to most closely represent this definition thus in this document, analysis of the ITT plus group, rather than the ITT group, is presented. Results of the efficacy analyses in the ITT group are largely consistent with those of the ITT plus group. (Section 3.1.2)
- Reviewer analyses and sponsor analyses numerically differ by a very small amount. The sponsor presents results using the Markov Chain Monte Carlo (MCMC) simulation citing that as per the statistical analysis plan, the outcome of the MCMC analyses were provided (in lieu of the more traditional use of the normal approximation methods) because the distributions for sensitivity and specificity were not absolutely normal. However, with the large size of this study, it is unclear how the distributions of sensitivities and specificities could be dramatically non-normal. Therefore, this review provides results of the more traditional normal approximation methods, presenting, among other results, the 95% confidence intervals for the differences in sensitivities and specificities calculated based on McNemar's test for paired data. Given that the normal approximation methods were the primary methods specified for the efficacy analysis in the protocol and statistical analysis plan and that the distributions of the sensitivities and specificities in a large study should be adequately normal, the normal approximation methods are preferred and are presented in Table 1. Qualitative conclusions resulting from the two approaches are largely the same. (Section 3.1.2)
- Analyses addressing the primary efficacy objectives are presented under varying conditions. Since it was unclear in the protocol whether the ITT plus or PP group was considered primary, analyses in both groups are presented. Analyses implementing varying cutoffs for the methacholine challenge (16 mg/mL, the standard published in the ATS guidelines, 12 mg/mL, as specified in the statistical analysis plan, and 4 mg/mL due to interest from the FDA medical review team) are presented. In general, a worst case

approach was used for addressing missing data; however, an analysis ignoring the missing data is also presented. In general, none of these criteria dramatically impacted the results of the analyses. (Sections 3.1.1 and 3.1.2)

- Although two of the three a priori documented study objectives were not met, the efficacy of aridol for regulatory purposes may still be substantiated by this study in that, it may be argued that the primary efficacy objectives defined as part of this study are not the most relevant in terms of assessing the efficacy of the aridol challenge test for regulatory approval. (Section 3.1.2)
- Prior to NDA submission, the Division expressed an interest in demonstrating that the sensitivity and specificity of aridol challenge are similar to that of a comparator procedure. This requires definition of the clinical meaning of *similarity* in sensitivity and specificity. As this study was not designed with this noninferiority objective in mind, no a priori noninferiority margin was documented in the protocol. In the absence of such documentation, we use the confidence intervals for the differences between aridol and methacholine in sensitivity and specificity to simply illustrate the degree to which the two diagnostic tests are the same and leave to clinical judgment whether this level of precision is acceptable in order to conclude that the two procedures are providing analogous levels of information. (Section 3.1.2)
- At the request of the FDA medical review team, plots of the cumulative dose of aridol or methacholine by the mean percent change from baseline in FEV_1 for the exercise positive and exercise negative strata are provided in this review. However, the use of these plots may be limited since the *mean* percent change from baseline in FEV_1 may be a misleading endpoint in this setting. First, the mean result may not be a good indicator for what will happen to a typical individual subject. And second, since subjects with the greatest falls in FEV_1 at the lower cumulative doses do not proceed to the higher cumulative doses (as they are diagnosed as positive and dosing stops), the impact of missing data on the mean fall in FEV_1 becomes more pronounced for the higher cumulative doses. In lieu of examining the *mean* percent change from baseline in FEV_1, plots of the percent change from baseline in FEV_1 for each individual could be considered. Such plots are provided in Appendix II. (Section 3.1.2 and Appendix II)
- The primary efficacy analyses by age, gender, and race are provided. No differing treatment effects among the subgroups examined were noted. (Section 4.1 and Appendix I)

5.2 Conclusions and Recommendations

Study 305 adequately demonstrates that aridol provides better diagnostic ability than chance alone, that is, the sensitivity and specificity of the aridol test exceed 50%. The confidence intervals for the differences between aridol and methacholine in sensitivity and specificity illustrate the degree to which the two diagnostic tests are the same and can be used to make a judgment regarding whether the two test are providing noninferior levels of information. While the estimates of the differences in sensitivity and specificity between aridol and methacholine are numerically small, it is important to consider these differences in context of the small benefit over chance alone.

6. APPENDIX I

Table 1a: Calculation of Sensitivity and Specificity of Mannitol and Methacholine Relative to Exercise with comparison between Mannitol and Methacholine **

		Exercise						Exercise		
		Positive	Negative	Row Total				Positive	Negative	Row Total
Aridol	Positive	101	90	191		Methacholine	Positive	93	79	172
	Negative	74	154	228			Negative	82	165	247
	Column Total	175	244	419*			Column Total	175	244	419*
Sensitivity (95% CI)		101/175=58% (50%, 65%)				Sensitivity (95% CI)		93/175=53% (46%, 61%)		
Specificity (95% CI)		154/244=63% (57%, 69%)				Specificity (95% CI)		165/244=68% (62%, 73%)		
By-Treatment-Group Difference (95% CI)										
Sensitivity (95% CI)		5% (-4%, 13%)								
Specificity (95% CI)		-5% (-12%, 3%)								

* n=419 since first exercise challenge is missing (and second exercise challenge is negative) for subject 28003

** ITT plus analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 16, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 1b: Direct comparison of Mannitol and Methacholine to assess concordance and discordance for exercise positive and exercise negative groups**

Exercise Positive Subjects		Methacholine			Exercise Negative Subjects		Methacholine		
		Positive	Negative	Row Total			Positive	Negative	Row Total
Aridol	Positive	67 (38%)	34 (19%)	101	Aridol	Positive	44 (18%)	46 (19%)	90
	Negative	26 (15%)	48 (27%)	74		Negative	35 (14%)	119 (49%)	154
	Column Total	93	82	175*		Column Total	79	165	244*

* n=419 since first exercise challenge is missing (and second exercise challenge is negative) for subject 28003

** ITT plus analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 16, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 2a: Calculation of Sensitivity and Specificity of Mannitol and Methacholine Relative to Exercise with comparison between Mannitol and Methacholine **

		Exercise						Exercise		
		Positive	Negative	Row Total				Positive	Negative	Row Total
Aridol	Positive	101	87	188		Methacholine	Positive	93	78	171
	Negative	72	153	225			Negative	80	162	242
	Column Total	173	240	413*			Column Total	173	240	413*
Sensitivity (95% CI)		101/173=58% (51%, 66%)				Sensitivity (95% CI)		93/173=54% (46%, 61%)		
Specificity (95% CI)		153/240=64% (58%, 70%)				Specificity (95% CI)		162/240=68% (62%, 73%)		
By-Treatment-Group Difference (95% CI)										
Sensitivity (95% CI)		5% (-4%, 13%)								
Specificity (95% CI)		-4% (-11%, 3%)								

* subjects in the ITT plus group with at least one diagnosis or the exercise standard of truth missing were ignored
** ITT plus analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 16, and missing data ignored

Table 2b: Direct comparison of Mannitol and Methacholine to assess concordance and discordance for exercise positive and exercise negative groups

Exercise Positive Subjects		Methacholine			Exercise Negative Subjects		Methacholine		
		Positive	Negative	Row Total			Positive	Negative	Row Total
Aridol	Positive	67 (39%)	34 (20%)	101	Aridol	Positive	43 (18%)	44 (18%)	87
	Negative	26 (9%)	46 (27%)	72		Negative	35 (15%)	118 (49%)	153
	Column Total	93	80	173*		Column Total	78	162	240*

* subjects in the ITT plus group with at least one diagnosis or the exercise standard of truth missing were ignored
** ITT plus analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 16, and missing data ignored

Table 3a: Calculation of Sensitivity and Specificity of Mannitol and Methacholine Relative to Exercise with comparison between Mannitol and Methacholine **

		Exercise						Exercise		
		Positive	Negative	Row Total				Positive	Negative	Row Total
Aridol	Positive	101	90	191		Methacholine	Positive	88	68	157
	Negative	74	154	228			Negative	87	176	262
	Column Total	175	244	419*			Column Total	175	244	419*
Sensitivity (95% CI)		101/175=58% (50%, 65%)				Sensitivity (95% CI)		88/175=50% (43%, 58%)		
Specificity (95% CI)		154/244=63% (57%, 69%)				Specificity (95% CI)		176/244=72% (67%, 78%)		
By-Treatment-Group Difference (95% CI)										
Sensitivity (95% CI)		7% (-2%, 16%)								
Specificity (95% CI)		-9% (-16%, -2%)								

* n=419 since first exercise challenge is missing (and second exercise challenge is negative) for subject 28003

** ITT plus analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 12, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 3b: Direct comparison of Mannitol and Methacholine to assess concordance and discordance for exercise positive and exercise negative groups

Exercise Positive Subjects		Methacholine			Exercise Negative Subjects		Methacholine		
		Positive	Negative	Row Total			Positive	Negative	Row Total
Aridol	Positive	62 (35%)	39 (22%)	101	Aridol	Positive	40 (16%)	50 (20%)	90
	Negative	26 (15%)	48 (27%)	74		Negative	28 (11%)	126 (52%)	154
	Column Total	88	87	175*		Column Total	68	176	244*

* n=419 since first exercise challenge is missing (and second exercise challenge is negative) for subject 28003

** ITT plus analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 12, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 4a: Calculation of Sensitivity and Specificity of Mannitol and Methacholine Relative to Exercise with comparison between Mannitol and Methacholine **

		Exercise						Exercise		
		Positive	Negative	Row Total				Positive	Negative	Row Total
Aridol	Positive	101	90	191		Methacholine	Positive	62	39	101
	Negative	74	154	228			Negative	113	205	318
	Column Total	175	244	419*			Column Total	175	244	419*
Sensitivity (95% CI)		101/175=58% (50%, 65%)				Sensitivity (95% CI)		62/175=35% (28%, 43%)		
Specificity (95% CI)		154/244=63% (57%, 69%)				Specificity (95% CI)		205/244=84% (79%, 89%)		
By-Treatment-Group Difference (95% CI)										
Sensitivity (95% CI)		22% (14%, 31%)								
Specificity (95% CI)		-21% (-27%, -14%)								

* n=419 since first exercise challenge is missing (and second exercise challenge is negative) for subject 28003

** ITT plus analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 4, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 4b: Direct comparison of Mannitol and Methacholine to assess concordance and discordance for exercise positive and exercise negative groups

Exercise Positive Subjects		Methacholine			Exercise Negative Subjects		Methacholine		
		Positive	Negative	Row Total			Positive	Negative	Row Total
Aridol	Positive	47 (27%)	54 (31%)	101	Aridol	Positive	27 (11%)	63 (26%)	90
	Negative	15 (9%)	59 (34%)	74		Negative	12 (5%)	142 (58%)	154
	Column Total	62	113	175*		Column Total	39	205	244*

* n=419 since first exercise challenge is missing (and second exercise challenge is negative) for subject 28003

** ITT plus analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 4, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 5a: Calculation of Sensitivity and Specificity of Mannitol and Methacholine Relative to Exercise with comparison between Mannitol and Methacholine **

		Exercise						Exercise		
		Positive	Negative	Row Total				Positive	Negative	Row Total
Aridol	Positive	95	75	170		Methacholine	Positive	90	66	156
	Negative	68	137	205			Negative	73	146	219
	Column Total	163	212	375			Column Total	163	212	375
Sensitivity (95% CI)		95/163=58% (51%, 66%)				Sensitivity (95% CI)		90/163=55% (48%, 63%)		
Specificity (95% CI)		137/212=65% (58%, 71%)				Specificity (95% CI)		146/212=69% (63%, 75%)		
By-Treatment-Group Difference (95% CI)										
Sensitivity (95% CI)		3% (-6%, 12%)								
Specificity (95% CI)		-4% (-12%, 3%)								

** PP analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 16, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 5b: Direct comparison of Mannitol and Methacholine to assess concordance and discordance for exercise positive and exercise negative groups**

Exercise Positive Subjects		Methacholine			Exercise Negative Subjects		Methacholine		
		Positive	Negative	Row Total			Positive	Negative	Row Total
Aridol	Positive	67 (41%)	28 (17%)	95	Aridol	Positive	38 (18%)	37 (17%)	75
	Negative	23 (14%)	45 (28%)	68		Negative	28 (13%)	109 (51%)	137
	Column Total	90	73	163		Column Total	66	146	212

** PP analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 16, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 6a: Calculation of Sensitivity and Specificity of Mannitol and Methacholine Relative to Exercise with comparison between Mannitol and Methacholine **

		Exercise						Exercise		
		Positive	Negative	Row Total				Positive	Negative	Row Total
Aridol	Positive	95	75	170		Methacholine	Positive	85	56	141
	Negative	68	137	205			Negative	78	156	234
	Column Total	163	212	375			Column Total	163	212	375
Sensitivity (95% CI)		95/163=58% (51%, 66%)				Sensitivity (95% CI)		85/163=52% (44%, 60%)		
Specificity (95% CI)		137/212=65% (58%, 71%)				Specificity (95% CI)		156/212=74% (68%, 80%)		
By-Treatment-Group Difference (95% CI)										
Sensitivity (95% CI)		6% (-3%, 15%)								
Specificity (95% CI)		-9% (-16%, -2%)								

* n=419 since first exercise challenge is missing (and second exercise challenge is negative) for subject 28003

** PP analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 12, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 6b: Direct comparison of Mannitol and Methacholine to assess concordance and discordance for exercise positive and exercise negative groups**

Exercise Positive Subjects		Methacholine			Exercise Negative Subjects		Methacholine		
		Positive	Negative	Row Total			Positive	Negative	Row Total
Aridol	Positive	62 (38%)	33 (20%)	95	Aridol	Positive	34 (16%)	41 (19%)	75
	Negative	23 (14%)	45 (28%)	68		Negative	22 (10%)	115 (54%)	137
	Column Total	85	78	163		Column Total	56	156	212

* n=419 since first exercise challenge is missing (and second exercise challenge is negative) for subject 28003

** PP analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 12, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 7a: Calculation of Sensitivity and Specificity of Mannitol and Methacholine Relative to Exercise with comparison between Mannitol and Methacholine in Subjects 6 to 11 years of age **

		Exercise						Exercise		
		Positive	Negative	Row Total				Positive	Negative	Row Total
Aridol	Positive	14	8	22		Methacholine	Positive	15	10	25
	Negative	7	7	14			Negative	6	5	11
	Column Total	21	15	36			Column Total	21	15	36
Sensitivity (95% CI)		14/21=67% (47%, 87%)				Sensitivity (95% CI)		15/21=71% (52%, 91%)		
Specificity (95% CI)		7/15=47% (21%, 72%)				Specificity (95% CI)		5/15=33% (9%, 57%)		
By-Treatment-Group Difference (95% CI)										
Sensitivity (95% CI)		-5% (-29%, 20%)								
Specificity (95% CI)		17% (-29%, 62%)								

** ITT analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 16, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 7b: Direct comparison of Mannitol and Methacholine to assess concordance and discordance for exercise positive and exercise negative groups in Subjects 6 to 11 years of age**

Exercise Positive Subjects		Methacholine			Exercise Negative Subjects		Methacholine		
		Positive	Negative	Row Total			Positive	Negative	Row Total
Aridol	Positive	11 (52%)	3 (14%)	14	Aridol	Positive	2 (17%)	3 (25%)	5
	Negative	4 (19%)	3 (14%)	7		Negative	5 (42%)	2 (17%)	7
	Column Total	15	6	21		Column Total	7	5	12

** ITT analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 16, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 8a: Calculation of Sensitivity and Specificity of Mannitol and Methacholine Relative to Exercise with comparison between Mannitol and Methacholine in Subjects 6 to 11 years of age **

		Exercise						Exercise		
		Positive	Negative	Row Total				Positive	Negative	Row Total
Aridol	Positive	14	8	22		Methacholine	Positive	14	9	23
	Negative	7	7	14			Negative	7	6	13
	Column Total	21	15	36			Column Total	21	15	36
Sensitivity (95% CI)		14/21=67% (47%, 87%)				Sensitivity (95% CI)		14/21=67% (47%, 87%)		
Specificity (95% CI)		7/15=47% (21%, 72%)				Specificity (95% CI)		6/15=40% (15%, 65%)		
By-Treatment-Group Difference (95% CI)										
Sensitivity (95% CI)		0% (-26%, 26%)								
Specificity (95% CI)		7% (-32%, 46%)								

** ITT analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 12, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 8b: Direct comparison of Mannitol and Methacholine to assess concordance and discordance for exercise positive and exercise negative groups in Subjects 6 to 11 years of age**

Exercise Positive Subjects		Methacholine			Exercise Negative Subjects		Methacholine		
		Positive	Negative	Row Total			Positive	Negative	Row Total
Aridol	Positive	10 (48%)	4 (19%)	14	Aridol	Positive	4 (%)	4 (%)	8
	Negative	4 (19%)	3 (14%)	7		Negative	5 (%)	2 (%)	7
	Column Total	14	7	21		Column Total	9	6	15

** ITT analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 12, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 9a: Calculation of Sensitivity and Specificity of Mannitol and Methacholine Relative to Exercise with comparison between Mannitol and Methacholine in Subjects 12 to 17 years of age **

		Exercise						Exercise		
		Positive	Negative	Row Total				Positive	Negative	Row Total
Aridol	Positive	17	15	32		Methacholine	Positive	20	14	34
	Negative	14	24	38			Negative	11	25	36
	Column Total	31	39	70			Column Total	31	39	70
Sensitivity (95% CI)		17/31=55% (37%, 72%)				Sensitivity (95% CI)		20/31=65% (48%, 81%)		
Specificity (95% CI)		24/39=62% (46%, 77%)				Specificity (95% CI)		25/39=64% (49%, 79%)		
By-Treatment-Group Difference (95% CI)										
Sensitivity (95% CI)		-10% (-32%, 13%)								
Specificity (95% CI)		-3% (-24%, 19%)								

** ITT analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 16, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 9b: Direct comparison of Mannitol and Methacholine to assess concordance and discordance for exercise positive and exercise negative groups in Subjects 12 to 17 years of age**

Exercise Positive Subjects		Methacholine			Exercise Negative Subjects		Methacholine		
		Positive	Negative	Row Total			Positive	Negative	Row Total
Aridol	Positive	12 (39%)	5 (16%)	17	Aridol	Positive	5 (13%)	10 (26%)	15
	Negative	8 (26%)	6 (19%)	14		Negative	9 (23%)	15 (38%)	24
	Column Total	20	11	31		Column Total	14	25	39

** ITT analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 16, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 10a: Calculation of Sensitivity and Specificity of Mannitol and Methacholine Relative to Exercise with comparison between Mannitol and Methacholine in Subjects 17+ years of age **

		Exercise						Exercise		
		Positive	Negative	Row Total				Positive	Negative	Row Total
Aridol	Positive	17	15	32		Methacholine	Positive	20	10	30
	Negative	14	24	38			Negative	11	29	40
	Column Total	31	39	70			Column Total	31	39	70
Sensitivity (95% CI)		17/31=55% (37%, 72%)				Sensitivity (95% CI)		20/31=65% (48%, 81%)		
Specificity (95% CI)		24/39=62% (46%, 77%)				Specificity (95% CI)		29/39=74% (61%, 88%)		
By-Treatment-Group Difference (95% CI)										
Sensitivity (95% CI)		-10% (-32%, 13%)								
Specificity (95% CI)		-13% (-32%, 6%)								

** ITT analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 12, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 10b: Direct comparison of Mannitol and Methacholine to assess concordance and discordance for exercise positive and exercise negative groups in Subjects 17+ years of age**

Exercise Positive Subjects		Methacholine			Exercise Negative Subjects		Methacholine		
		Positive	Negative	Row Total			Positive	Negative	Row Total
Aridol	Positive	12 (39%)	5 (16%)	17	Aridol	Positive	5 (13%)	10 (26%)	15
	Negative	8 (26%)	6 (19%)	14		Negative	5 (13%)	19 (49%)	24
	Column Total	20	11	31		Column Total	10	29	39

** ITT analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 12, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 11a: Calculation of Sensitivity and Specificity of Mannitol and Methacholine Relative to Exercise with comparison between Mannitol and Methacholine in Subjects 17+ years of age **

		Exercise						Exercise		
		Positive	Negative	Row Total				Positive	Negative	Row Total
Aridol	Positive	73	69	142		Methacholine	Positive	60	57	117
	Negative	56	127	183			Negative	69	139	208
	Column Total	129	196	325			Column Total	129	196	325
Sensitivity (95% CI)		73/129=57% (48%, 65%)				Sensitivity (95% CI)		60/129=47% (38%, 55%)		
Specificity (95% CI)		127/196=65% (58%, 71%)				Specificity (95% CI)		139/196=71% (65%, 77%)		
By-Treatment-Group Difference (95% CI)										
Sensitivity (95% CI)		10% (1%, 20%)								
Specificity (95% CI)		-6% (-14%, 1%)								

** ITT analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 16, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 11b: Direct comparison of Mannitol and Methacholine to assess concordance and discordance for exercise positive and exercise negative groups in Subjects 17+ years of age**

Exercise Positive Subjects		Methacholine			Exercise Negative Subjects		Methacholine		
		Positive	Negative	Row Total			Positive	Negative	Row Total
Aridol	Positive	46 (36%)	27 (21%)	73	Aridol	Positive	34 (17%)	35 (18%)	69
	Negative	14 (11%)	42 (33%)	56		Negative	23 (12%)	103 (53%)	126
	Column Total	60	69	129		Column Total	57	138	195

** ITT analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 16, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 12a: Calculation of Sensitivity and Specificity of Mannitol and Methacholine Relative to Exercise with comparison between Mannitol and Methacholine in Subjects 17+years of age **

		Exercise						Exercise		
		Positive	Negative	Row Total				Positive	Negative	Row Total
Aridol	Positive	73	69	142		Methacholine	Positive	56	51	107
	Negative	56	127	183			Negative	73	145	218
	Column Total	129	196	325			Column Total	129	196	325
Sensitivity (95% CI)		73/129=57% (48%, 65%)				Sensitivity (95% CI)		56/129=44% (35%, 52%)		
Specificity (95% CI)		127/196=65% (58%, 71%)				Specificity (95% CI)		145/196=74% (68%, 80%)		
By-Treatment-Group Difference (95% CI)										
Sensitivity (95% CI)		13% (3%, 23%)								
Specificity (95% CI)		-10% (-17%, -2%)								

** ITT analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 12, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 12b: Direct comparison of Mannitol and Methacholine to assess concordance and discordance for exercise positive and exercise negative groups in Subjects 17+ years of age**

Exercise Positive Subjects		Methacholine			Exercise Negative Subjects		Methacholine		
		Positive	Negative	Row Total			Positive	Negative	Row Total
Aridol	Positive	42 (33%)	31 (24%)	73	Aridol	Positive	31 (16%)	39 (20%)	70
	Negative	14 (11%)	42 (33%)	56		Negative	20 (10%)	106 (54%)	126
	Column Total	56	73	129		Column Total	51	145	196

** ITT analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 12, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 13a: Calculation of Sensitivity and Specificity of Mannitol and Methacholine Relative to Exercise with comparison between Mannitol and Methacholine in Males **

		Exercise						Exercise		
		Positive	Negative	Row Total				Positive	Negative	Row Total
Aridol	Positive	46	44	90		Methacholine	Positive	38	36	74
	Negative	34	73	107			Negative	43	81	124
	Column Total	80	117	197			Column Total	81	117	198
Sensitivity (95% CI)		58% (47%, 68%)				Sensitivity (95% CI)		47% (36%, 58%)		
Specificity (95% CI)		62% (54%, 71%)				Specificity (95% CI)		69% (61%, 78%)		
By-Treatment-Group Difference (95% CI)										
Sensitivity (95% CI)		10% (-3%, 22%)								
Specificity (95% CI)		-7% (-17%, 3%)								

** ITT analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 16, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 13b: Direct comparison of Mannitol and Methacholine to assess concordance and discordance for exercise positive and exercise negative groups in Males**

Exercise Positive Subjects		Methacholine			Exercise Negative Subjects		Methacholine		
		Positive	Negative	Row Total			Positive	Negative	Row Total
Aridol	Positive	28 (35%)	18 (22%)	46	Aridol	Positive	22 (19%)	22 (19%)	44
	Negative	10 (12%)	25 (31%)	35		Negative	14 (12%)	59 (50%)	73
	Column Total	38	43	81		Column Total	36	81	117

** ITT analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 16, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 14a: Calculation of Sensitivity and Specificity of Mannitol and Methacholine Relative to Exercise with comparison between Mannitol and Methacholine in Males **

		Exercise						Exercise		
		Positive	Negative	Row Total				Positive	Negative	Row Total
Aridol	Positive	46	44	90		Methacholine	Positive	37	34	71
	Negative	34	73	107			Negative	44	83	127
	Column Total	80	117	197			Column Total	81	117	198
Sensitivity (95% CI)		58% (47%, 68%)				Sensitivity (95% CI)		46% (35%, 57%)		
Specificity (95% CI)		62% (54%, 71%)				Specificity (95% CI)		71% (63%, 79%)		
By-Treatment-Group Difference (95% CI)										
Sensitivity (95% CI)		11% (-2%, 24%)								
Specificity (95% CI)		-9% (-19%, 2%)								

** ITT analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 12, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 14b: Direct comparison of Mannitol and Methacholine to assess concordance and discordance for exercise positive and exercise negative groups in Males**

Exercise Positive Subjects		Methacholine			Exercise Negative Subjects		Methacholine		
		Positive	Negative	Row Total			Positive	Negative	Row Total
Aridol	Positive	27 (33%)	19 (23%)	46	Aridol	Positive	20 (17%)	24 (21%)	44
	Negative	10 (12%)	25 (31%)	35		Negative	14 (19%)	59 (50%)	73
	Column Total	37	44	81		Column Total	34	83	117

** ITT analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 12, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 15a: Calculation of Sensitivity and Specificity of Mannitol and Methacholine Relative to Exercise with comparison between Mannitol and Methacholine in Females **

		Exercise						Exercise		
		Positive	Negative	Row Total				Positive	Negative	Row Total
Aridol	Positive	55	46	101		Methacholine	Positive	55	43	98
	Negative	39	81	120			Negative	39	84	123
	Column Total	94	127	221			Column Total	94	127	221
Sensitivity (95% CI)		59% (49%, 68%)				Sensitivity (95% CI)		59% (49%, 68%)		
Specificity (95% CI)		64% (55%, 72%)				Specificity (95% CI)		66% (58%, 74%)		
By-Treatment-Group Difference (95% CI)										
Sensitivity (95% CI)		0% (-12%, 12%)								
Specificity (95% CI)		-2% (-13%, 8%)								

** ITT analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 16, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 15b: Direct comparison of Mannitol and Methacholine to assess concordance and discordance for exercise positive and exercise negative groups in Females**

Exercise Positive Subjects		Methacholine			Exercise Negative Subjects		Methacholine		
		Positive	Negative	Row Total			Positive	Negative	Row Total
Aridol	Positive	39 (41%)	16 (17%)	55	Aridol	Positive	22 (17%)	24 (19%)	46
	Negative	16 (17%)	23 (24%)	39		Negative	21 (17%)	60 (47%)	81
	Column Total	55	39	94		Column Total	43	84	127

** ITT analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 16, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 16a: Calculation of Sensitivity and Specificity of Mannitol and Methacholine Relative to Exercise with comparison between Mannitol and Methacholine in Females **

		Exercise						Exercise		
		Positive	Negative	Row Total				Positive	Negative	Row Total
Aridol	Positive	55	46	101		Methacholine	Positive	51	34	85
	Negative	39	81	120			Negative	43	93	136
	Column Total	94	127	221			Column Total	94	127	221
Sensitivity (95% CI)		59% (49%, 68%)				Sensitivity (95% CI)		54% (44%, 64%)		
Specificity (95% CI)		64% (55%, 72%)				Specificity (95% CI)		73% (66%, 81%)		
By-Treatment-Group Difference (95% CI)										
Sensitivity (95% CI)		4% (-8%, 17%)								
Specificity (95% CI)		-9 (-19%, 0%)								

** ITT analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 12, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 16b: Direct comparison of Mannitol and Methacholine to assess concordance and discordance for exercise positive and exercise negative groups in Females**

Exercise Positive Subjects		Methacholine			Exercise Negative Subjects		Methacholine		
		Positive	Negative	Row Total			Positive	Negative	Row Total
Aridol	Positive	35 (37%)	20 (21%)	55	Aridol	Positive	20 (16%)	26 (20%)	46
	Negative	16 (17%)	23 (24%)	39		Negative	14 (11%)	67 (53%)	81
	Column Total	51	43	94		Column Total	34	93	127

** ITT analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 12, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 17a: Calculation of Sensitivity and Specificity of Mannitol and Methacholine Relative to Exercise with comparison between Mannitol and Methacholine in Caucasians **

		Exercise						Exercise		
		Positive	Negative	Row Total				Positive	Negative	Row Total
Aridol	Positive	75	69	144		Methacholine	Positive	75	61	136
	Negative	54	117	171			Negative	57	125	182
	Column Total	129	186	315			Column Total	132	186	318
Sensitivity (95% CI)		58% (50%, 67%)				Sensitivity (95% CI)		57% (48%, 65%)		
Specificity (95% CI)		63% (56%, 70%)				Specificity (95% CI)		67% (60%, 74%)		
By-Treatment-Group Difference (95% CI)										
Sensitivity (95% CI)		2% (-8%, 12%)								
Specificity (95% CI)		-4% (-12%, 4%)								

** ITT analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 16, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 17b: Direct comparison of Mannitol and Methacholine to assess concordance and discordance for exercise positive and exercise negative groups in Caucasians**

Exercise Positive Subjects		Methacholine			Exercise Negative Subjects		Methacholine		
		Positive	Negative	Row Total			Positive	Negative	Row Total
Aridol	Positive	52 (40%)	23 (18%)	75	Aridol	Positive	36 (19%)	33 (18%)	69
	Negative	20 (16%)	34 (26%)	54		Negative	25 (13%)	92 (49%)	117
	Column Total	72	57	129		Column Total	61	125	186

** ITT analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 16, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 18a: Calculation of Sensitivity and Specificity of Mannitol and Methacholine Relative to Exercise with comparison between Mannitol and Methacholine in Caucasians **

		Exercise						Exercise		
		Positive	Negative	Row Total				Positive	Negative	Row Total
Aridol	Positive	75	69	144		Methacholine	Positive	67	51	118
	Negative	54	117	171			Negative	62	135	197
	Column Total	129	186	315			Column Total	129	186	315
Sensitivity (95% CI)		58% (50%, 67%)				Sensitivity (95% CI)		52% (43%, 61%)		
Specificity (95% CI)		63% (56%, 70%)				Specificity (95% CI)		73% (66%, 79%)		
By-Treatment-Group Difference (95% CI)										
Sensitivity (95% CI)		6% (-4%, 17%)								
Specificity (95% CI)		-10% (-17%, -2%)								

** ITT analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 12, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 18b: Direct comparison of Mannitol and Methacholine to assess concordance and discordance for exercise positive and exercise negative groups in Caucasians**

Exercise Positive Subjects		Methacholine			Exercise Negative Subjects		Methacholine		
		Positive	Negative	Row Total			Positive	Negative	Row Total
Aridol	Positive	47 (36%)	28 (22%)	75	Aridol	Positive	33 (18%)	36 (19%)	69
	Negative	20 (16%)	34 (26%)	54		Negative	18 (10%)	99 (53%)	117
	Column Total	67	62	129		Column Total	51	135	186

** ITT analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 12, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 19a: Calculation of Sensitivity and Specificity of Mannitol and Methacholine Relative to Exercise with comparison between Mannitol and Methacholine in Non-Caucasians **

		Exercise						Exercise		
		Positive	Negative	Row Total				Positive	Negative	Row Total
Aridol	Positive	26	21	47		Methacholine	Positive	21	18	39
	Negative	20	37	57			Negative	25	40	65
	Column Total	46	58	104			Column Total	46	58	104
Sensitivity (95% CI)		57% (42%, 71%)				Sensitivity (95% CI)		46% (31%, 60%)		
Specificity (95% CI)		64% (51%, 76%)				Specificity (95% CI)		69% (57%, 81%)		
By-Treatment-Group Difference (95% CI)										
Sensitivity (95% CI)		11% (-6%, 28%)								
Specificity (95% CI)		-5% (-21%, 11%)								

** ITT analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 16, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 19b: Direct comparison of Mannitol and Methacholine to assess concordance and discordance for exercise positive and exercise negative groups in Non-Caucasians**

Exercise Positive Subjects		Methacholine			Exercise Negative Subjects		Methacholine		
		Positive	Negative	Row Total			Positive	Negative	Row Total
Aridol	Positive	15 (33%)	11 (24%)	26	Aridol	Positive	8 (14%)	13 (22%)	21
	Negative	6 (13%)	14 (30%)	20		Negative	10 (17%)	27 (47%)	37
	Column Total	21	25	46		Column Total	18	40	58

** ITT analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 16, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 20a: Calculation of Sensitivity and Specificity of Mannitol and Methacholine Relative to Exercise with comparison between Mannitol and Methacholine in Non-Caucasians **

		Exercise						Exercise		
		Positive	Negative	Row Total				Positive	Negative	Row Total
Aridol	Positive	26	21	47		Methacholine	Positive	21	17	38
	Negative	20	37	57			Negative	25	41	66
	Column Total	46	58	104			Column Total	46	58	104
Sensitivity (95% CI)		57% (42%, 71%)				Sensitivity (95% CI)		46% (31%, 60%)		
Specificity (95% CI)		64% (51%, 76%)				Specificity (95% CI)		71% (59%, 82%)		
By-Treatment-Group Difference (95% CI)										
Sensitivity (95% CI)		11% (-6%, 28%)								
Specificity (95% CI)		-7% (-23%, 10%)								

** ITT analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 12, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 20b: Direct comparison of Mannitol and Methacholine to assess concordance and discordance for exercise positive and exercise negative groups in Non-Caucasians**

Exercise Positive Subjects		Methacholine			Exercise Negative Subjects		Methacholine		
		Positive	Negative	Row Total			Positive	Negative	Row Total
Aridol	Positive	15 (33%)	11 (24%)	26	Aridol	Positive	7 (12%)	14 (24%)	21
	Negative	6 (13%)	14 (30%)	20		Negative	10 (17%)	27 (47%)	37
	Column Total	21	25	46		Column Total	17	41	58

** ITT analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 12, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

Table 21a: Calculation of Sensitivity and Specificity of Mannitol and Methacholine Relative to Physician Diagnosis with comparison between Mannitol and Methacholine **

		Physician Diagnosis						Physician Diagnosis		
		Positive	Negative	Row Total				Positive	Negative	Row Total
Aridol	Positive	139	50	189		Methacholine	Positive	128	45	173
	Negative	118	112	230			Negative	129	117	246
	Column Total	257	162	419			Column Total	257	162	419
Sensitivity (95% CI)		54% (48%, 60%)				Sensitivity (95% CI)		50% (44%, 56%)		
Specificity (95% CI)		69% (62%, 76%)				Specificity (95% CI)		72% (65%, 79%)		
By-Treatment-Group Difference (95% CI)										
Sensitivity (95% CI)		4% (-3%, 11%)								
Specificity (95% CI)		-3% (-12%, 6%)								

* n=419 since first exercise challenge is missing (and second exercise challenge is negative) for subject 28003

** ITT plus analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 16, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

*** Physician Diagnosis of "probably", "possible", "very likely", and "extremely likely or definite" considered positive. "Unlikely but not excluded" and "very unlikely or excluded" considered negative.

Table 21b: Direct comparison of Mannitol and Methacholine to assess concordance and discordance for Physician Diagnosed Positive and Negative groups**

Physician Diagnosed Positive Subjects		Methacholine			Physician Diagnosed Negative Subjects		Methacholine		
		Positive	Negative	Row Total			Positive	Negative	Row Total
Aridol	Positive	89 (35%)	50 (19%)	139	Aridol	Positive	21 (13%)	29 (18%)	50
	Negative	39 (15%)	79 (31%)	118		Negative	24 (15%)	88 (54%)	112
	Column Total	128	129	257		Column Total	45	117	162

* n=419 since first exercise challenge is missing (and second exercise challenge is negative) for subject 28003

** ITT plus analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 16, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

*** Physician Diagnosis of "probably", "possible", "very likely", and "extremely likely or definite" considered positive. "Unlikely but not excluded" and "very unlikely or excluded" considered negative.

Table 22a: Calculation of Sensitivity and Specificity of Mannitol and Methacholine Relative to Physician Diagnosis with comparison between Mannitol and Methacholine

		Physician Diagnosis						Physician Diagnosis		
		Positive	Negative	Row Total				Positive	Negative	Row Total
Aridol	Positive	139	50	189		Methacholine	Positive	116	41	157
	Negative	118	112	230			Negative	141	121	262
	Column Total	257	162	419			Column Total	257	162	419
Sensitivity (95% CI)		54% (48%, 60%)				Sensitivity (95% CI)		45% (39%, 51%)		
Specificity (95% CI)		69% (62%, 76%)				Specificity (95% CI)		75% (68%, 81%)		
By-Treatment-Group Difference (95% CI)										
Sensitivity (95% CI)		9% (2%, 16%)								
Specificity (95% CI)		-10% (-25%, 5%)								

* n=419 since first exercise challenge is missing (and second exercise challenge is negative) for subject 28003

** ITT plus analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 12, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

*** Physician Diagnosis of "probable", "possible", "very likely", and "extremely likely or definite" considered positive. "Unlikely but not excluded" and "very unlikely or excluded" considered negative.

Table 22b: Direct comparison of Mannitol and Methacholine to assess concordance and discordance for physician diagnosed positive and physician diagnosed negative groups

Physician Diagnosed Positive Subjects		Methacholine			Physician Diagnosed Negative Subjects		Methacholine		
		Positive	Negative	Row Total			Positive	Negative	Row Total
Aridol	Positive	81 (32%)	58 (23%)	139	Aridol	Positive	20 (12%)	30 (19%)	50
	Negative	35 (14%)	83 (32%)	118		Negative	21 (13%)	91 (56%)	112
	Column Total	116	141	257		Column Total	41	121	162

* n=419 since first exercise challenge is missing (and second exercise challenge is negative) for subject 28003

** ITT plus analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 12, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

*** Physician Diagnosis of "probably", "possible", "very likely", and "extremely likely or definite" considered positive. "Unlikely but not excluded" and "very unlikely or excluded" considered negative.

Table 23a: Calculation of Sensitivity and Specificity of Mannitol and Methacholine Relative to Physician Diagnosis with comparison between Mannitol and Methacholine **

		Physician Diagnosis						Physician Diagnosis		
		Positive	Negative	Row Total				Positive	Negative	Row Total
Aridol	Positive	132	37	169		Methacholine	Positive	122	34	156
	Negative	108	98	206			Negative	118	101	219
	Column Total	240	135	375			Column Total	240	135	375
Sensitivity (95% CI)		55% (49%, 61%)				Sensitivity (95% CI)		51% (45%, 57%)		
Specificity (95% CI)		73% (65%, 80%)				Specificity (95% CI)		75% (67%, 82%)		
By-Treatment-Group Difference (95% CI)										
Sensitivity (95% CI)		4% (-3%, 11%)								
Specificity (95% CI)		-2% (-11%, 7%)								

* n=419 since first exercise challenge is missing (and second exercise challenge is negative) for subject 28003
** PP analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 16, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)
*** Physician Diagnosis of "probably", "possible", "very likely", and "extremely likely or definite" considered positive. "Unlikely but not excluded" and "very unlikely or excluded" considered negative.

Table 23b: Direct comparison of Mannitol and Methacholine to assess concordance and discordance for Physician Diagnosed Positive and Negative groups**

Physician Diagnosed Positive Subjects		Methacholine			Physician Diagnosed Negative Subjects		Methacholine		
		Positive	Negative	Row Total			Positive	Negative	Row Total
Aridol	Positive	88 (37%)	44 (18%)	132	Aridol	Positive	16 (12%)	21 (16%)	37
	Negative	34 (14%)	74 (31%)	108		Negative	18 (13%)	80 (59%)	98
	Column Total	122	118	240		Column Total	34	101	135

* n=419 since first exercise challenge is missing (and second exercise challenge is negative) for subject 28003
** PP plus analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 16, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)
*** Physician Diagnosis of "probably", "possible", "very likely", and "extremely likely or definite" considered positive. "Unlikely but not excluded" and "very unlikely or excluded" considered negative.

Table 24a: Calculation of Sensitivity and Specificity of Mannitol and Methacholine Relative to Physician Diagnosis with comparison between Mannitol and Methacholine

		Physician Diagnosis						Physician Diagnosis		
		Positive	Negative	Row Total				Positive	Negative	Row Total
Aridol	Positive	132	37	169		Methacholine	Positive	110	31	141
	Negative	108	98	206			Negative	130	104	234
	Column Total	240	135	375			Column Total	240	135	375
Sensitivity (95% CI)		55% (49%, 61%)				Sensitivity (95% CI)		46% (40%, 52%)		
Specificity (95% CI)		73% (65%, 80%)				Specificity (95% CI)		77% (70%, 84%)		
By-Treatment-Group Difference (95% CI)										
Sensitivity (95% CI)		9% (2%, 16%)								
Specificity (95% CI)		-4% (-13%, 4%)								

* n=419 since first exercise challenge is missing (and second exercise challenge is negative) for subject 28003

** PP plus analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 12, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

*** Physician Diagnosis of "probably", "possible", "very likely", and "extremely likely or definite" considered positive. "Unlikely but not excluded" and "very unlikely or excluded" considered negative.

Table 24b: Direct comparison of Mannitol and Methacholine to assess concordance and discordance for physician diagnosed positive and physician diagnosed negative groups

Physician Diagnosed Positive Subjects		Methacholine			Physician Diagnosed Negative Subjects		Methacholine		
		Positive	Negative	Row Total			Positive	Negative	Row Total
Aridol	Positive	80 (33%)	52 (22%)	132	Aridol	Positive	15 (11%)	22 (16%)	37
	Negative	30 (13%)	78 (33%)	108		Negative	16 (12%)	82 (61%)	98
	Column Total	110	130	240		Column Total	31	104	135

* n=419 since first exercise challenge is missing (and second exercise challenge is negative) for subject 28003

** PP plus analysis group, outcome of aridol challenge with subjects with 10% between dose fall in FEV1 considered positive, methacholine cutoff of 12, and worst case for missing data (when missing, aridol assumed to be wrong and methacholine assumed to be correct)

*** Physician Diagnosis of "probably", "possible", "very likely", and "extremely likely or definite" considered positive. "Unlikely but not excluded" and "very unlikely or excluded" considered negative.

7. APPENDIX II

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Application Type/Number	Submission Type/Number	Submitter Name	Product Name
NDA-22368	ORIG-1	PHARMAXIS LTD	ARIDOL POWDER FOR INHALATION

This is a representation of an electronic record that was signed electronically and this page is the manifestation of the electronic signature.

/s/

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12/03/2009

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