



BC Centre for Disease Control
AN AGENCY OF THE PROVINCIAL HEALTH SERVICES AUTHORITY

Investigation of adverse events after intra-vitreous
injection of Avastin™,
Canada, October – December 2008

5 December 2008 – 24 February 2009

British Columbia Centre for Disease Control

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1. INTRODUCTION

A cluster of adverse events, namely sterile endophthalmitis /Toxic Anterior Segment Syndrome (TASS) was associated with the intra-vitreous injection of a specific packaging lot of bevacizumab (Avastin™) administered for the treatment of Age-related Macular Degeneration (AMD).

While indicated for the treatment of metastatic colon cancer, intra-vitreous injection of Avastin has been used off label for the treatment of AMD. The reported baseline incidence of sterile endophthalmitis in this setting ranges up to 2%[1]. An increase was reported by one ophthalmology clinic in British Columbia starting in October, 2008. Twenty-two of 195 patients (11%) developed an AR after treatment with Avastin. A cohort study conducted by University of British Columbia (UBC) and British Columbia Centre for Disease Control (BCCDC) found an association with a specific package lot of Avastin (B3002B028). Subsequently, 6 other Canadian centres reported cases.

Field Epidemiologist assistance was requested by the BCCDC to assist with additional case finding and data management from other ophthalmology clinics reporting cases as well as cases that may have been reported to Health Canada's Marketed Health Products Directorate (MHPD) in the Health Products and Foods Branch. This report summarizes the investigation to date and concludes with some recommendations.

2. BACKGROUND

Macular degeneration is the most common cause of severe vision loss in Canada, particularly among the elderly [2]. There are two types of macular degeneration, wet and dry. The wet form of AMD (neovascular) occurs when abnormal blood vessels develop under the retina. These new blood vessels tend to be very fragile and often leak blood and fluid, damaging the retina. Only 10% of AMD patients have this form of the disease, but they account for more than 90% of patients with severe vision loss.

Promising treatments for neovascular AMD thus involve drugs that inhibit the growth of new blood vessels. Pharmacologic therapies initially included verteporfin photodynamic therapy and pegaptanib sodium, however they were only moderately effective – slowing progression but only rarely improving vision [3,4]. Ranibizumab (Lucentis™), an anti-vascular endothelial growth factor (anti-VEGF) monoclonal antibody fragment, was developed specifically for the treatment of neovascular AMD. Prior to the availability of Lucentis, however, ophthalmologists began using Avastin, a full length anti-VEGF monoclonal antibody closely related to ranibizumab [5,6]. When Lucentis was approved in 2006, its high cost of over \$1,500 per dose and need for multiple injections over months to years, precluded easy replacement of Avastin despite the availability of an approved drug. To date, Lucentis is only funded in a few provinces. Avastin is now the focus of a large multi-centre trial underway in the United States sponsored by the National Institutes of Health, to compare it with Lucentis [7]. Final results from this study will not be available until 2011, but first year data may be available later this year.



3. METHODS

Confirmed case definitions

Vancouver

A patient who developed an adverse reaction (which included Sterile Endophthalmitis/TASS, inflammation, blurred vision or floaters) after intra-vitreous injection of Avastin for Age-related Macular Degeneration administered by an ophthalmologist in Vancouver between 1 October – 27 October 2008.

Toronto

A patient who developed an adverse reaction (which included Sterile Endophthalmitis/TASS, inflammation, blurred vision or floaters) after intra-vitreous injection of Avastin for Age-related Macular Degeneration administered by an ophthalmologist in Toronto between 20 October – 29 October 2008.

Calgary

A patient who developed an adverse reaction (which included Sterile Endophthalmitis/TASS, inflammation, blurred vision or floaters) after intra-vitreous injection of Avastin for Age-related Macular Degeneration administered by an ophthalmologist in Calgary between 1 October – 3 December 2008.

Case finding

Vancouver and Toronto

All patients from the Vancouver and Toronto clinic who received an injection during the time period of concern (1 October – 27 October 2008: Vancouver; 20 October – 29 October 2008: Toronto) were contacted and asked to come to the clinic for evaluation. Enhanced passive surveillance was carried out after the end of the time period of concern.

Calgary

Only patients who reported an ADR to the clinic were followed up and identified as cases.



Data Collection

Data were collected using a standardized questionnaire and entered into a Microsoft Access database (Appendix A).

Analytic Epidemiology

Vancouver and Toronto

All patients who received an injection during the time period of interest were included in the study.

Calgary

Non-cases were selected by reviewing electronic patient charts of the cases and selecting up to 2 patients who received an injection by the same surgeon within a day or two of the treatment date of the case.

Standardized information was abstracted from patients' charts in all clinic centres.

Data Analysis

Logistic regression was used to determine how the risk of ADR related to potential risk factors. Univariate models were fit for each risk factor. To explore various multivariate models and ultimately choose a final model, all demographic variables and their two-way interactions were used in stepwise selection. To assess whether any variables in the final model were subject to confounding by any variables that had been omitted from the final model, each omitted variable was re-introduced individually. The impact on the sign, magnitude, and significance of each of the original coefficients was examined.

Cases and non-cases were compared with regards to age and sex. Differences between means were T-tested. Due to the differences in case finding techniques, Calgary's data were analyzed separately from Vancouver and Toronto's data.

All statistical analyses were carried out in SAS version 9.1.

Laboratory testing

Product samples

Samples from lot B3002B028 and B33437 were analyzed by mass spectrometer at UBC Laboratory. Samples obtained from the manufacturer were tested for LAL.



Patient samples

Samples were also obtained from several patients in both clinics and cultured.

4. RESULTS

Descriptive Epidemiology

Vancouver

Between 9 and 27 October 2008 a total of 21 confirmed cases out of approximately 151 (14%) patients were reported from six surgeons in Vancouver (Figure 1). The mean age of confirmed cases was 71 years (median, 74 years; range, 45-90 years) eleven (52%) of 21 were female. Among the 21 cases, adverse reactions included blurred vision/floaters 21 (100%), inflammation 13 (62%), and sterile endophthalmitis 7 (33%). Two patients reported their ADR to the clinic. The remaining 19 patients were identified through active case finding and patient follow up by the clinic. There were no reports of hospitalization or serious complication.

Between 9 to 27 October 2008 the Vancouver clinic used Lot B3002B028 and B33437. On 13 November 2008, the clinic began using Lot B3002B018. There has only been one reported ADR since the use of the implicated lot was discontinued.

Toronto

A total of 6 of 6 (100%) patients who received Avastin injections between Oct 20 and 29th 2008 experienced an ADR. Age-related macular degeneration was the underlying diagnosis for 1 (17%) of 6 confirmed cases (Table 1). The mean age of confirmed cases was 68 years (median, 67 years; range, 51 to 84 years). Three (50%) of 6 confirmed cases were female. Adverse reactions included; blurred vision/floaters 6 (100%), inflammation 4 (67%), and elevated intraocular pressure 3 (50%). All patients received Lot B3002B028.

Calgary

Between 7 October and 3 December 2008 a total of 21 adverse reactions out of approximately 1080 (2%) patients, were reported from four surgeons in Calgary (Figure 1). Age-related macular degeneration was the underlying diagnosis for the majority 18 (86%) of confirmed cases (Table 1). The mean age of confirmed cases was 74 years (median, 77 years; range, 27 to 90 years). Eleven (52%) of 21 confirmed cases were female. Adverse reactions included; blurred vision/floaters 19 (90%), inflammation 21 (100%), and pain 9 (43%). All 21 patients who reported their ADR to the clinic were symptomatic. There were no reports of hospitalization or serious complication.



The Calgary clinic reported that they used Lot B3002B028 from 1 October to 3 December 2008 and began using B3002B018 on 4 December 2008. All patients with reported ADRs between 7 October and 3 December 2008 received Lot B3002B028. This clinic treats between 100-150 patients (with Avastin) per week. They have had one ADR reported in January with the new lot (B3002B018).

Figure 1: Reported adverse events meeting the case definition by Lot number and date of injection, Calgary, Toronto and Vancouver clinics, 5 October – 12 December 2008 (n=48)

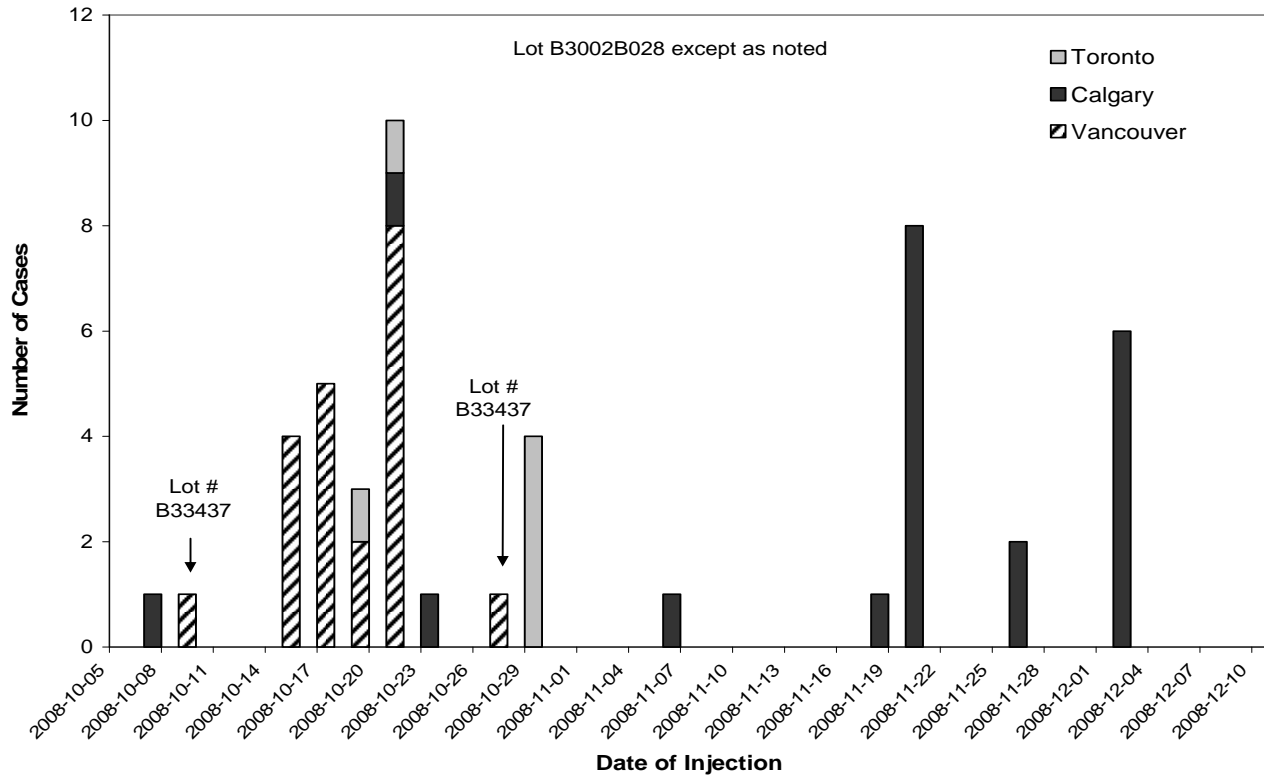




Table 1: Age and sex demographics of cases by clinic, Calgary, Toronto and Vancouver (n=48)

	Calgary	Toronto	Vancouver
No. (%) female	11 (52%)	3 (50%)	11 (52%)
Mean age	74	68	71
Median age	77	67	74
Age range	27-90	51-84	45-90

Table 2: Underlying Diagnosis/ reason for treatment with Avastin for cases by clinic (n=48)

Underlying Diagnosis	Calgary	Toronto	Vancouver
Age-related Macular Degeneration	18	1	9
Choroidal neovascularisation	0	0	3
Cystoid macular edema	0	1	5
DM Retinopathy	2	2	2
Other*	1	2	2
Total	21	6	21

*Other includes: a patient with both Central Retinal Vein Occlusion and Cystoid Macular Edema and a patient with both Age-related Macular Degeneration and Choroidal neovascularisation



Analytical Epidemiology

Univariate analysis

Vancouver and Toronto

Results of the univariate analysis are shown in Table 3. The risk of ADR was significantly associated with the lot number patient received ($p < 0.0001$), number of prior injections ($p = 0.0166$) and underlying diagnosis ($p = 0.0196$) but not with age ($p = 0.2494$) or sex ($p = 0.1654$).

The odds of ADR for patients who received Lot B3002B028 were 29 times higher than patients who received Lot B33437 ($p < 0.0001$). The injection variable was grouped into both 3 and 4 levels and analyzed. The grouping with 3 levels was statistically significant ($p = 0.0166$) in the univariate analysis and therefore included in the multivariate analysis.

No statistically significant differences were found between cases and non-cases with regards to age and sex.



Table 3: Odds ratios (OR), 95% confidence interval (CI) for odds ratios and P-values from the univariate analysis of the relationship between risk factors and adverse reactions in patients of two clinic centres, Vancouver and Toronto (2008) (n=157)

Variable	OR	CI for OR	P-value
Number of prior injections (3 levels)			0.0166
0 ^a	1.00	-	-
1-2	0.39	0.10-1.58	0.1885
3 or more	2.03	0.68-6.07	0.2076
Number of prior injections (4 levels)			0.0226
0 ^a	1.00	-	-
1-2	0.39	0.10-1.58	0.1885
3-6	1.65	0.51-5.31	0.4011
7 or more	3.15	0.83-11.96	0.0918
Age			0.2494
	0.98	0.95-1.01	0.2432
Sex			0.1632
Male ^a	1.00	-	-
Female	0.55	0.24-1.27	0.1632
Lot number			<0.0001
B33437 ^a	1.00	-	-
B3002B028	29.2	6.58-129.2	<0.0001
Underlying diagnosis			0.0196
AMD ^a	1.00	-	-
Non-AMD	2.75	1.18-6.42	0.0196

^a reference group

Calgary

Although Calgary's data were not statistically significant, they did show the same trend regarding number of prior injections as well as the specific lot number as Vancouver's data. Results of the univariate analyses are available in Appendix A.

Multivariate analysis

Vancouver and Toronto

The final multivariate model included number of prior injections, lot number, and underlying diagnosis. Reintroduction of the variables excluded from the final model (age and sex) did not impact the sign, magnitude, or significance of any of the coefficients for number of prior injections, lot number or underlying diagnosis. Therefore, number of prior injections, lot number and underlying diagnosis were the variables included in the final multivariate model (Table 4).



Even after adjusting for number of prior injections and underlying diagnosis, patients who received lot B3002B028 had 33 times greater odds of having an ADR than patients who did not receive the implicated lot.

Table 4: Adjusted odds ratios (OR), 95% confidence interval (CI) for odds ratios and P-values from the final multivariate model of the relationship between risk factors and ADRs in patients of two clinic centres Vancouver and Toronto (2008) (n=157)

Variable	Adjusted OR	CI for OR	P-value
Number of prior injections			0.0128
0 prior injections ^a	1.00	-	-
1-2 prior injections	0.58	0.12-2.88	0.5056
3 or more prior injections	3.70	0.91-15.02	0.0676
Lot number			<0.0001
B33437 ^a	1.00	-	-
B3002B028	32.9	7.04-154.01	<0.0001
Underlying Diagnosis			0.0240
AMD ^a	1.00	-	-
Non-AMD	3.60	1.18-10.92	0.0240

^a reference group

Laboratory results

All three lots used at the Vancouver clinic were tested at UBC Pharmacological Laboratory. No chemical differences were detected between the lots. Endotoxin testing also conducted at UBC on leftover syringes found that even after 3 months of storage, there was no detectable endotoxin.

Initial laboratory testing conducted by Health Canada found that endotoxin levels and pH were within the specifications for the product. A final analysis report is pending.

Drug preparation methods

Vancouver

Avastin for injection is prepared by a compounding pharmacy that draws up doses from a multi-dose vial through multiple punctures with individual syringes and needles. The syringes and empty multi-dose vial are then shipped to the clinic where the doses are stored refrigerated until use. Storage time of a syringe ranges from 2 days to no more than 2 weeks.



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Toronto

Avastin for injection is prepared by a compounding pharmacy that performs a single puncture of a multi-dose vial and draws up each dose in a separate syringe and adds a needle. Each vial can fill up to 21 syringes, which are then packaged together and sent to the clinic.

Calgary

The clinic receives boxes containing single multi-dose vials of Avastin from a local pharmacy. Each vial is punctured once and doses are drawn up into individual syringes, needles are attached and the syringes are all used the same day. All materials are kept refrigerated until use. As many as 2-3 vials are used in any given day.



5. DISCUSSION

This outbreak represents the largest cluster of ADRs associated with intra-vitreous injections of Avastin reported in Canada. Forty-eight patients from 3 provinces reported an adverse reaction meeting the case definition in the fall of 2008. The importance of this investigation is evident in the large number of patients who are dependent on this off-label drug for treatment of wet AMD, given the otherwise prohibitive cost of the approved product to many patients.

The analytic epidemiology identified a higher risk of ADR among patients who received Lot B3002B028. However there were no significant findings from the lab results to explain what may have caused the ADR. Further testing of the implicated lot continues in an effort to identify the cause of the ADRs. There has been speculation as to whether it was a formulation issue or a preparation/storage/ administration issue. The formulation issue is complicated as this particular lot was part of a larger batch and that the difference between the implicated lot (B3002B028) and another lot (B3002B018) was only in the packaging of the pre-filled vials for delivery from the manufacturer.

Patients who received three or more prior injections appear to have a greater chance of developing an ADR after adjusting for lot number and underlying diagnosis, than those patients who did not have any prior injections. While this apparent association was not statistically significant ($p=0.068$) in our study, this finding is consistent with other case studies of ADRs where patients who experienced an ADR were more likely to have had previous injections [1]. They hypothesized that with increased sensitization there may be an increased immune response. Further exploration is required to understand the association between number of prior injections and ADR and if this phenomenon was enhanced with the implicated lot.

Although univariate analysis of the Calgary data was not statistically significant, it did show the same trend as Vancouver and Toronto's data. The inability to achieve significance may be due to the potential of cases being represented among the non-cases (since non-reports were considered to have had no ADR), since likely the patients who reported ADRs were more severe. Whereas Vancouver and Toronto ADRs were identified by active case finding and follow up with all patients who received an injection, therefore we can be more confident that the cases and non-cases are representative of true cases and true non-cases and therefore provide a better estimate of the association.

The need for representative baseline data was identified during the investigation. Although the number of ADRs reported during the time period of the investigation appeared higher than expected, there was little information available which quantified the number of ADRs reported prior to the investigation. Standardizing how patients are deemed to have had an ADR as well as maintaining a count of the number of injections of Avastin administered, will provide a better baseline in the future.



During this investigation, it became apparent that drug preparation practices differed among the 3 clinic centres. Practices varied from a compounding pharmacy preparing the drug to preparation at the clinic. This lack of standardization however, did not appear to have affected the findings but may need further exploration.

6. RECOMMENDATIONS

- To enable detection of any concerns that may arise from a single lot of Avastin or other clinical practice issue, consistently document injection technique such as syringe and needle type and size along with the lot number on patient charts. The latter to include as appropriate both the packaging lot number found on the box and lot number from the product vial.
- To better document adverse events following the use of Avastin, including baseline rates in order to detect future outbreaks, a protocol for prospective surveillance should be developed and executed. This protocol will need to have a clear case definition for ADRs, and record patient demographics, product lot numbers and relevant clinical details.



7. ACKNOWLEDGEMENTS

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9. APPENDICES

List of documents included in Appendix:

- a. Additional tables
- b. Data collection tools
 - i. Patient Form
 - ii. Clinic Form



a. Additional tables – comparison of cases and control

Table 3: Calgary and Vancouver cases and non-cases by age and sex

	Calgary		Vancouver	
	Cases (n=21)	Non-cases (n=29)	Cases (n=21)	Non-cases (n=130)
No. (%) female	11 (52%)	13 (44%)	11 (52%)	86 (66%)
Mean age	74	78	71	74
Median age	77	79	74	76
Age range	27-90	56-92	45-90	34-94

Table 4: Calgary univariate analysis

Variable	OR	CI	P-value
Injection (3 levels)			0.2519
0	1.00	-	-
1-2	0.67	0.04-10.25	0.3961
3 or more	2.13	0.18-25.78	0.2197
Injection (4 levels)			0.3860
0	1.00	-	-
1-2	0.67	0.04-10.25	0.2636
3-6	2.57	0.19-34.47	0.2076
7 or more	1.78	0.13-23.52	0.5681
Age			0.1921
	0.96	0.90-1.02	0.2249
Sex			0.5977
Male	1.00	-	-
Female	1.35	0.44-4.18	0.5981
Underlying Diagnosis			0.9025
Non-AMD	1.00	-	-
AMD	1.13	0.17-7.45	0.9028
Lot number			0.2600
B3002B028 (only one lot)	-	-	-



b. Data collection tools

Patient Form: Investigation of adverse events after treatment with Avastin™ for age-related macular degeneration, Canada

Complete this form for each patient who received treatment with Avastin between Oct and Dec 2008

Patient

Patient ID: _____ Patient Initials: _____ Clinic ID: _____

Date of Birth (YY/MM/DD): _____ or Age: _____ Sex: M F

Underlying Diagnosis/ Reason for treatment with Avastin™ (check all that apply):

- Age-related macular degeneration
- Cystoid macular edema
- Central retinal vein occlusion
- DM retinopathy
- Other Specify: _____
- Choroidal neovascularisation
- Branch retinal vein occlusion
- Diabetic macular edema

Drug Information

Date of Injection (YY/MM/DD): _____ Eye(s): OD OS

Lot number: (shipping box/vial):

Box Vial

- B3002B028 / B30028
- B3002B018 / B30028
- B33437 / B33437
- Other Specify: _____

Number of prior injections of Avastin™ patient has received: _____

Date of most recent prior dose (YY/MM/DD): _____



Adverse Reaction(s) (check all that apply):

- No Reaction
- Inflammation
- Floaters
- Other details, Specify: _____
- TASS
- Sterile endophthalmitis
- Blurred Vision

Symptomatic: Yes No If symptomatic; Date of Symptom Onset (YY/MM/DD): _____

Date of Diagnosis (YY/MM/DD): _____

Management of ADR investigation:

- Lab testing, specify type and result: _____
- Treatment: _____

