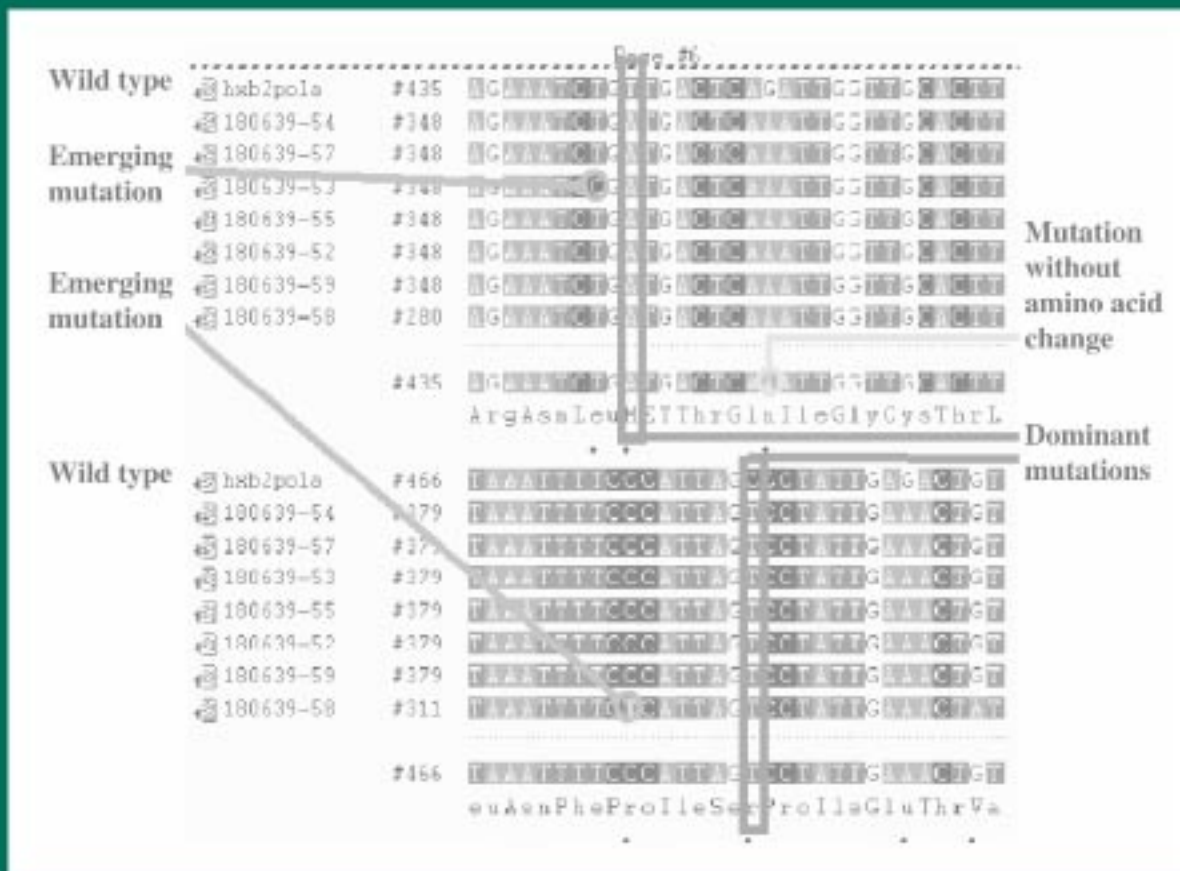




HIV-1 Strain and Primary Drug Resistance in Canada

Surveillance Report to June 30, 2002



December 2002

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to June 30, 2002**

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Division of HIV/AIDS Epidemiology and Surveillance
Division of Retrovirus Surveillance
National HIV Laboratories
Centre for Infectious Disease Prevention and Control
Population and Public Health Branch
Health Canada

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Acknowledgements: We acknowledge the provincial and territorial HIV/AIDS coordinators, laboratories, health care providers and reporting physicians for providing sera and non-nominal, confidential epidemiologic data that enabled this report to be published. We also acknowledge the Scientific Publication and Multimedia Services, Population and Public Health Branch, Health Canada, for its contribution in editing and producing the report both for printing and posting on the Internet.

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Suggested citation: Health Canada. *HIV-1 Strain and Primary Drug Resistance in Canada. Surveillance Report to June 30, 2002*. Division of HIV/AIDS Epidemiology and Surveillance, Centre for Infectious Disease Prevention and Control, Health Canada, 2002.

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Cat # H39-625/2003

ISBN # 0-662-67325-5

(On-line) ISSN 0-662-33970-3

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The Division of HIV/AIDS Epidemiology and Surveillance and the national HIV laboratories at the Centre for Infectious Disease Prevention and Control, Health Canada, are pleased to provide you with the *HIV-1 Strain and Primary Drug Resistance in Canada: Surveillance Report to June 30, 2002*. Primary drug resistance is a term used to identify resistance observed in HIV-infected individuals who have never before received treatment and so presumably have been infected with a drug-resistant strain of HIV.

This report presents data that are shared by the provinces/territories participating in the Canadian HIV Strain and Drug Resistance Surveillance Program. The Division of HIV/AIDS Epidemiology and Surveillance is responsible for data management, data analyses, and writing and coordination of the publication of this report. The Division of Retrovirus Surveillance is responsible for coordinating the collection of the HIV data. The National Laboratory for HIV Genetics is responsible for conducting the subtyping and drug resistance genotyping.

The major finding in this surveillance report is that primary drug resistance to antiretroviral drugs has been identified in 7.1% of our sample population of 847 treatment naïve individuals. Resistance to more than one class of antiretroviral drug has been identified in 0.7% of the sample population. For the first time, we report primary drug resistance to non-nucleoside reverse transcriptase inhibitors. Primary drug resistance has been observed in females and males; across different age groups, ethnicities, and exposure categories; in HIV-1 subtype A, B, and C infections; and in recent and prevalent HIV infections. With respect to HIV-1 subtypes, subtype B continues to predominate in Canada, 93.1% of the samples belonging to this group, but HIV-1 subtypes A, C, D, and E and the recombinant subtypes A/B, A/C, and A/G have been identified across Canada. Significantly higher proportions of non-B infections were detected among females (compared with males), among people reporting heterosexual contact as their primary exposure factor, and among people of Black, Asian or mixed ethnicities. There is geographic variation in the prevalence of non-B HIV-1 subtypes, and this variation is likely related to travel and migration from countries where other subtypes predominate.

From a public health perspective, while primary drug resistance may not yet be a significant problem in Canada, it will no doubt play a significant role in shaping the course of the HIV epidemic in this country and worldwide. For this reason, the data from the Canadian HIV Strain and Drug Resistance Surveillance Program will be valuable to guide Canada's response to the public health problems posed by drug-resistant HIV, including our efforts to develop treatment strategies and effective prevention programs. The introduction of variant HIV-1 subtypes into Canada also requires vigilant monitoring in order to ensure that diagnostic and screening algorithms can adequately detect all circulating strains and to inform vaccine research and development.

This is the second report of results from the Canadian HIV Strain and Drug Resistance Surveillance Program. We will be working towards improving this report to reflect changes in the surveillance of HIV strains and drug resistance. We welcome and appreciate your comments and suggestions.

Yours sincerely,



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Table of Contents

| | |
|----------------------------------------------------------------------------------------------------------------------------------------------------------------------|----------|
| Results at a Glance | 1 |
| 1.0 Introduction | 3 |
| 1.1 An overview of the Canadian HIV Strain and Drug Resistance Surveillance Program | 3 |
| 1.2 Goals of the CHSDRSP | 3 |
| 2.0 Methodology | 5 |
| 2.1 Epidemiologic data and laboratory specimen collection and transfer | 5 |
| 2.2 Genetic algorithm for HIV subtyping and drug resistance testing | 5 |
| 2.3 Consensus of major mutations associated with primary drug resistance | 5 |
| 2.4 Epidemiologic analyses | 6 |
| 3.0 HIV-1 Primary Drug Resistance (1997-2001) | 7 |
| 3.1 Background | 7 |
| 3.2 Laboratory tests to detect drug resistance | 7 |
| 3.3 Data sources | 7 |
| 3.4 Prevalence and determinants of HIV-1 primary drug resistance in the sample population (<i>N</i> = 847) | 8 |
| Table 1 Prevalence of primary drug resistance among treatment naïve individuals with newly diagnosed infection (1997-2001) | 8 |
| Table 2 Major mutations identified in HIV-1 reverse transcriptase and protease | 9 |
| Table 3 Prevalence of drug resistance among treatment naïve individuals with newly diagnosed infection by year of diagnosis | 10 |
| Table 4 Prevalence of drug resistance among treatment naïve individuals with newly diagnosed infection by province | 11 |
| Table 5 Prevalence of drug resistance among treatment naïve individuals with newly diagnosed infection by age at diagnosis | 12 |
| Table 6 Prevalence of drug resistance among treatment naïve individuals with newly diagnosed infection by gender | 12 |
| Table 7 Prevalence of drug resistance among treatment naïve individuals with newly diagnosed infection by exposure category | 13 |
| Table 8 Prevalence of drug resistance among treatment naïve individuals with newly diagnosed infection by ethnicity | 14 |
| Table 9 Prevalence of drug resistance among treatment naïve individuals by HIV-1 subtype | 15 |
| Table 10 Prevalence of drug resistance among treatment naïve individuals by recently acquired versus prevalent HIV-1 infection | 15 |
| Table 11 Summary of key studies on drug resistance among treatment naïve individuals with newly diagnosed infection in Canada | 16 |
| Table 12 Summary of key studies on drug resistance among treatment naïve individuals with newly diagnosed infection in the United States and in Western Europe | 17 |

| | |
|---------------------------------------------------------------------------------------------------------------|-----------|
| 4.0 HIV-1 Subtypes (1984-January 2002) | 19 |
| 4.1 Background | 19 |
| 4.2 Data sources | 19 |
| 4.3 Prevalence and determinants of HIV-1 subtypes in sample population (<i>N</i> = 1,312) | 20 |
| Table 13 Prevalence of HIV-1 subtypes among treatment naïve individuals with newly diagnosed infection | 20 |
| Table 14 Prevalence of HIV-1 subtypes by year of diagnosis with HIV-1 infection | 21 |
| Table 15 Prevalence of HIV-1 subtypes by province | 21 |
| Table 16 Prevalence of HIV-1 subtypes by age of diagnosis with HIV-1 infection | 22 |
| Table 17 Prevalence of HIV-1 subtypes by gender | 22 |
| Table 18 Prevalence of HIV-1 subtypes by exposure category | 23 |
| Table 19 Prevalence of HIV-1 subtypes by ethnicity | 24 |
| Table 20 Prevalence of HIV-1 subtypes by recently acquired versus prevalent HIV-1 infections | 24 |
| Table 21 Prevalence of HIV-1 subtypes by primary drug resistance | 25 |
| Table 22 Epidemiologic characteristics of individuals infected with non-B subtypes of HIV-1 | 26 |
| | |
| 5.0 National HIV Reference Services | 29 |
| 5.1 Background | 29 |
| Table 23 HIV-1 subtype distribution of samples submitted to reference services | 29 |
| 5.2 HIV-2 infections | 30 |
| | |
| Technical Notes | 31 |
| | |
| Data Limitations | 33 |
| | |
| Appendix 1 Glossary of terms | 35 |
| | |
| Appendix 2 List of major mutations used for this report and corresponding antiretroviral drugs | 37 |

Results at a Glance

This report is organized into five sections. The first section provides an overview of Health Canada's Canadian HIV Strain and Drug Resistance Surveillance Program (CHSDRSP). The second section describes the methodologies used for data collection, transfer and analyses. The third section describes the primary drug resistance results from the program. This section also summarizes results from other key studies conducted in Canada, the United States, and Western Europe. The fourth section describes HIV-1 subtype results from the program. The fifth section describes the sentinel surveillance arm of the CHSDRSP, which serves the provincial public health laboratories by testing samples from individuals with unusual clinical manifestations and/or with unusual laboratory results for HIV subtype and drug resistance (if requested).

This part of the report provides a summary of the main findings:

- ◆ The overall prevalence of primary drug resistance to at least one antiretroviral drug is 7.1% in the sample population of 847 individuals with newly diagnosed HIV infection who have never received treatment.
- ◆ Multi-drug resistance (protease inhibitors/nucleoside reverse transcriptase inhibitors and non-nucleoside reverse transcriptase inhibitors/nucleoside reverse transcriptase inhibitors) has been identified in six treatment naïve individuals (0.7%) with newly diagnosed HIV in the sample population.
- ◆ Resistance to nucleoside reverse transcriptase inhibitors has been observed in cases with newly diagnosed HIV infection in this sample population as early as 1998; resistance to non-nucleoside reverse transcriptase inhibitors and protease inhibitors as well as multi-drug resistance have also been observed in such cases.
- ◆ Primary drug resistance has been observed in females and males; across different age groups, ethnicities, and exposure categories; in HIV-1 subtype A, B, and C infections; and in recent and prevalent HIV infections.
- ◆ In Canada, HIV-1 subtype B continues to predominate, 93.1% of the samples subtyped belonging to this group, but subtypes A, C, D, E (also known as the circulating recombinant A/E), recombinant A/B, recombinant A/C, and recombinant A/G have been identified across Canada
- ◆ There is geographic variation with respect to the prevalence of non-B HIV-1 subtypes. This variation is likely related to travel and migration from countries where other subtypes predominate.
- ◆ Significantly higher proportions of non-B infections were detected among females (compared with males), among people reporting heterosexual contact as their primary exposure factor, and among people of Black, Asian or mixed ethnicities.

Public Health Implications

- ◆ The prevalence of primary drug resistance can be used to develop population-based recommendations for initial therapies (especially for pregnant women and for use in post-exposure prophylaxis).
- ◆ The extent to which drug-resistant strains of HIV are being transmitted can serve as an indicator to evaluate the effectiveness of prevention programs.
- ◆ HIV isolates from different populations and changes over time can be monitored to evaluate diagnostic and screening algorithms to ensure that all circulating strains are adequately detected.
- ◆ HIV subtype data can be used for the research and development of vaccines.
- ◆ Increased knowledge about HIV genetic diversity will be useful in monitoring the spread of HIV in Canada including assessing transmission patterns and disease progression.

1.0 Introduction

1.1 An overview of the Canadian HIV Strain and Drug Resistance Surveillance Program

The Canadian HIV Strain and Drug Resistance Surveillance Program (CHSDRSP) is a collaborative effort between the provinces and territories and the Centre for Infectious Disease Prevention and Control (CIDPC), Population and Public Health Branch, Health Canada. It forms a key component in a national system for the enhanced and integrated surveillance of HIV/AIDS, STD, emerging retroviruses and other sexually transmitted blood-borne pathogens. The program was initiated to characterize and monitor the genetic diversity of the HIV epidemic in Canada, addressing the concerns of affected communities, public health authorities, primary care physicians and researchers.

1.2 Goals of the CHSDRSP

A 1998 consensus workshop in Vancouver established the following goals for this program:

To enhance the safety of the blood supply

In order to ensure the safety of the blood supply, all HIV tests need to reliably detect the strains circulating in the country. The precedent for this goal was the discovery of HIV-2 and highly divergent group O strains of HIV-1, which required modification of some serologic screening tests by the addition of new antigens to ensure detection. The sentinel arm of the CHSDRSP, through the reference services of the national HIV laboratories, addresses this goal by testing samples with unusual serologic, polymerase chain reaction (PCR) or other virologic test results. This relation between the national HIV laboratories and the provincial public health laboratories also serves other programs, including quality assurance and the monitoring of diagnostic kits.

To inform vaccine development

One of the primary, public health related reasons for conducting a systematic surveillance of HIV genetic variability in Canada is to inform vaccine research and development. The majority of vaccines undergoing clinical trials have been developed using strains of HIV-1 subtype B (predominant in North America and Europe). However, the ability of these vaccines to elicit cross-subtype responses is unclear. It is therefore imperative to collect data on the prevalence and incidence of HIV subtypes and other determinants of subtype diversity in Canada in order to inform vaccine strategies, since the efficacy and effectiveness of vaccines may be subtype specific.

To assess genetic markers of HIV drug resistance

Although antiretroviral therapies have led to a reduction in HIV related morbidity and mortality in Canada, there is concern that their widespread use, increased number of treatment failures, and high HIV infection rates may result in increased transmission of drug-resistant virus. Indeed, studies have shown that transmission of drug-resistant virus is increasing. The CHSDRSP aims to address the prevalence of primary drug resistance and the variation of this prevalence over time, geographic area and population risk group. The resulting information can be used to develop treatment guidelines at the population level for initial therapeutic regimens and to develop more effective HIV prevention strategies, including the prevention of mother-to-child transmission.

To determine HIV transmission, pathogenesis and progression to HIV-related diseases

Although genetic analyses have been used to assess the spread of HIV globally, there is little consensus on whether differences in HIV subtype and mutations conferring drug resistance affect the rate of transmission, pathogenesis and HIV-related disease progression. Knowing the distribution of HIV variants in Canada, along with corresponding epidemiologic information, will help to address these questions. The public health implications of such findings, including prevention and treatment strategies, are of special interest.

2.0 Methodology

2.1 Epidemiologic data and laboratory specimen collection and transfer

The provincial partners in the CHSDRSP send serum samples taken for diagnostic testing from treatment naïve individuals with newly diagnosed HIV infection to CIDPC, Health Canada. Subtype analysis and primary drug resistance genotyping is conducted at the National Laboratory for HIV Genetics. Incidence testing is conducted at the National HIV Reference Laboratory. Samples with unusual laboratory results are sent through the sentinel arm of the CHSDRSP and are analyzed for subtype and drug resistance (if this testing is requested) at the National Laboratory for HIV Genetics.

For each submitted laboratory sample, non-nominal epidemiologic information is also sent to CIDPC. The data include information routinely collected on the national or provincial HIV case reporting forms and, where available, additional information that helps interpret the laboratory results, including treatment history, CD4 count and viral load at diagnosis, and previous HIV testing history. Epidemiologic analyses are conducted at the Division of HIV/AIDS Epidemiology and Surveillance.

As of June 30, 2002, British Columbia (BC), Alberta, Manitoba, Saskatchewan, Newfoundland and Labrador, and Nova Scotia have participated in CHSDRSP. The results presented in this report represent samples that CIDPC had received as of June 30, 2002, on which HIV subtype and drug resistance testing had been completed successfully. Serum samples have also been received from Ontario, but since these samples were received through the sentinel arm of the CHSDRSP the results are reported in Section 5, under National HIV Reference Services.

Samples and epidemiologic data continue to flow to Health Canada from participating provinces, and results from these analyses will be presented in future reports. Discussions are currently under way to expand collection of samples and epidemiologic data to the remaining provinces and territories.

2.2 Genetic algorithm for HIV subtyping and drug resistance testing

After extraction of the RNA and an initial one-step reverse transcriptase PCR, nested PCR amplification of the *pol* gene (which encodes for the HIV protease and reverse transcriptase enzymes) is performed using a combination of published and in-house group M consensus primers. The PCR product is directly sequenced with internal PCR primers on a Li-Cor 4200L automated sequencer. In this way, a complete double stranded sequence for the entire protease gene and the first 253 amino acids of reverse transcriptase is used to assess subtype and primary mutations associated with drug resistance. Of note is that between July 1998 and December 2000, the C2-V5 region (233 amino acids) of the envelope protein was used to assess HIV subtype. When PCR products sequence poorly, at least two additional amplification and sequencing attempts are made with increasing enhancement algorithms to sequence the PCR product. If this is not successful, a final effort is made to resolve the problem by cloning the PCR product and screening 10 to 12 clones per person.

2.3 Consensus of major mutations associated with primary drug resistance

Interpretation of results from genetic algorithms requires knowledge of the association between specific mutations and virologic response to antiretroviral drugs. The associations are often complex and not necessarily additive. Consensus drug resistance mutation lists have been published through database banks (e.g. Stanford University, <http://hivdb.stanford.edu/hiv> and the Los Alamos HIV Sequence Database, http://resdb.lanl.gov/Resist_DB/) and by expert committees on HIV drug resistance (e.g. International AIDS Society – USA Drug Resistance Mutations Group). However, even the experts do not always agree on these so-called “rules-based” algorithms.

For this report, major mutations identified in the protease and reverse transcriptase genes of HIV were defined by a consensus of listings reported by the International AIDS Society-USA Drug Resistance Mutations Group^{*} and by Stanford University. The major mutations that have been added to our consensus list since the last surveillance report of samples received to June 30, 2001, include I50L, D67N, Y115F, L210W, K219E/Q, L100I, V108I, Y188C/H/L, P225H, M230L and P236L. Please refer to Appendix 2 for the complete, current list of mutations associated with clinical resistance that was used for this report.

2.4 Epidemiologic analyses

Laboratory and epidemiologic data are linked using unique identifiers. Significant associations between primary drug resistance or HIV-1 non-B subtypes and epidemiologic characteristics of individuals in the sample population were assessed using the χ^2 test and where appropriate Fisher's exact test. Logistic regression analyses to further define independent factors associated with primary drug resistance and with non-B infections were conducted using SPSS 8.0TM (SPSS Inc. Chicago, IL). Independent variables that were examined included age at diagnosis of HIV infection, gender, exposure category, ethnicity, and year of diagnosis of HIV infection.

* International AIDS Society-USA Drug Resistance Mutations Group. *Drug Resistance Mutations in HIV-1*. Topics in HIV Medicine 2002;10(2):11-15.

3.0. HIV-1 Primary Drug Resistance (1997-2001)

3.1 Background

In any infected individual, the HIV population is made up of wild type and variant strains. The variants contain mutations in the viral genome that are a result of the rapid but relatively inaccurate replication of HIV. Under selective pressure (e.g. as a result of antiretroviral drug treatment), variants that are resistant to the drug are able to grow and become predominant in the viral population. For some drugs (e.g. non-nucleoside reverse transcriptase inhibitors), a single mutation is sufficient to confer drug resistance. Such a mutation is referred to as a “major” mutation. For other drugs, (e.g. protease inhibitors) a combination of mutations is often required to confer resistance. Such mutations are known as “minor” mutations. Of note is that most mutations are lethal or neutral, and the wild type strain usually dominates in the absence of selective pressure from drug therapy because it replicates more efficiently.

Drug resistance is often cited as a contributing factor to treatment failure. Drug resistance that is associated with individuals who are already receiving treatment and that is described in the context of treatment failure is commonly referred to as “secondary” drug resistance. A phenomenon that has received considerable attention recently is the transmission of drug-resistant HIV-1. This type of drug resistance, also called “primary” drug resistance, has been reported in individuals who have never before received treatment for HIV and so have presumably been infected with drug-resistant HIV. Primary drug resistance is becoming more widespread in most countries where antiretroviral therapy is used. People infected with drug-resistant variants of HIV may be at increased risk of drug failure despite never having received treatment. However, the prevalence of primary drug resistance and the variation of this prevalence over time, geographic area and population risk group are not well understood.

The data presented in this report include data on major mutations associated with drug resistance from samples that were received by the National Laboratory for HIV Genetics as of June 30, 2002, and represent individuals with HIV infection newly diagnosed between 1997 and 2001.

3.2 Laboratory tests to detect drug resistance

Two types of tests are used to detect drug resistance, genotypic and phenotypic. Genotypic tests provide information about the genetic makeup of the virus by identifying the mutations that are strongly associated with resistance. Phenotypic testing measures the ability of a virus to replicate in the presence of varying drug concentrations. While the methodologies for both tests are well established, each has its limitations, described in the section Data Limitations. Genotyping was used to identify the major drug resistance mutations described in this report. A detailed description of the laboratory tests is given in Section 2, Methodology.

3.3 Data sources

This section highlights the main findings from the CHSDRSP up to June 30, 2002. It is important to note that the results presented here represent individuals who sought testing, whose condition was properly diagnosed, and whose test results were reported as HIV positive. Furthermore, the results are based only on those individuals for whom sufficient sera, taken for the purposes of diagnostic testing, were available to send to the national HIV laboratories by June 30, 2002, and, of these samples, the subset for whom reverse transcriptase PCR amplification and sequencing were successful in identifying major mutations.

As of June 30, 2002, serum samples from 1,645 individuals with HIV newly diagnosed between 1997 and 2001 and corresponding non-nominal epidemiologic data received from BC, Alberta, Manitoba, Saskatchewan and Nova Scotia for drug resistance genotyping. While the goal of the CHSDRSP is to collect serum samples from all cases with newly diagnosed infection, the data presented in this report are a result of convenience sampling methods and may not be representative. As well, discussions are under way to expand the program to the remaining provinces and territories.

At the time of writing of this report (December 2002) the National Laboratory for HIV Genetics has analyzed a total of 1,189 samples for major mutations. Viral RNA had been successfully amplified from 847 (71.2%) of the serum samples. This level of success in amplifying virus from serum specimens will likely improve further as sample quality is enhanced and through the identification and use of various primer combinations for reverse transcriptase PCR amplification.

For this report, major mutations identified in the protease and reverse transcriptase genes of HIV were defined by a consensus of listings reported by the International AIDS Society-USA Drug Resistance Mutations Group*. Please refer to Appendix 2 for the complete, current list of mutations associated with clinical resistance that was used for this report.

3.4 Prevalence and determinants of HIV-1 primary drug resistance in the sample population (N = 847)

Table 1 shows the prevalence of primary drug resistance in the sample of individuals with newly diagnosed HIV infection between 1997 and 2001. Major mutations were present in 7.1% of the sample population of 847 of these treatment naïve individuals. Note that since none of the individuals had previously received treatment, they may have been infected with a drug-resistant strain of HIV-1. Major mutations associated with nucleoside reverse transcriptase inhibitors (NRTIs), non-nucleoside reverse transcriptase inhibitors (NNRTIs) and protease inhibitors were identified in 35 (4.1%), 4 (0.5%), and 15 (1.8%) of individuals in the sample population respectively. It is notable that a major mutation (L90M) associated with resistance to the protease inhibitors nelfinavir and saquinavir was identified in a sample from Ontario that was sent through the sentinel arm of the CHSDRSP. Six individuals in the sample population (0.7%) were infected with multi-drug resistant HIV-1 harbouring major mutations to NRTI and protease inhibitors or to NRTIs and NNRTIs. Multi-drug resistant HIV was also identified in three samples from Ontario that were sent through the sentinel arm of the CHSDRSP, but since these individuals had previously been receiving antiretroviral treatment, primary drug resistance could not be identified.

Table 1: Prevalence of primary drug resistance among treatment naïve individuals with newly diagnosed infection (1997-2001)

| Primary drug resistance | Frequency | Percentage |
|----------------------------------------|------------|------------|
| Wild type/minor mutations ¹ | 787 | 92.9 |
| NRTI ² | 35 | 4.1 |
| NNRTI ³ | 4 | 0.5 |
| Protease | 15 | 1.8 |
| MDR ⁴ | 6 | 0.7 |
| Total | 847 | 100 |

¹ Wild type indicates that no major mutations associated with drug resistance were identified. Minor mutations refers to genetic variables not associated with drug resistance.

² NRTI refers to nucleoside reverse transcriptase inhibitor.

³ NNRTI refers to non-nucleoside reverse transcriptase inhibitor.

⁴ MDR refers to multi-drug resistance and includes mutations in HIV-1 that are associated with resistance to any two of the three classes of antiretroviral drugs (NRTIs, NNRTIs, and protease inhibitors).

* International AIDS Society-USA Drug Resistance Mutations Group. *Drug Resistance Mutations in HIV-1*. Topics in HIV Medicine 2002;10(2):11-15.

Table 2: Major mutations identified in HIV-1 reverse transcriptase and protease

| Anti-retroviral drug | Number of individuals (%) | Major mutation(s)¹ |
|----------------------------------------|----------------------------------|--------------------------------------|
| NRTI², Total | 35 (100) | |
| AZT, d4T | 21 (60) | M41L ³ |
| AZT, d4T | 1 (2.8) | M41L, L210W |
| AZT, d4T | 2 (5.8) | K70R |
| AZT, d4T | 1 (2.8) | K219Q |
| AZT, d4T ddC | 1 (2.8) | M41L, T69D, T69N |
| AZT, d4T ddC | 1 (2.8) | T69N, K70R |
| AZT, d4T 3TC | 1 (2.8) | M184V, T215Y |
| ddC | 1 (2.8) | T69N |
| ddC | 2 (5.8) | T69D, T69N |
| Adefovir | 1 (2.8) | K70E |
| 3TC | 2 (5.8) | M184V |
| 3TC | 1 (2.8) | M184I, M184V |
| NNRTI⁴, Total | 4 (100) | |
| DLV, EFV | 1 (25) | G190A, G190E, G190S |
| DLV, EFV, NVP | 2 (50) | K103N |
| DLV, EFV, NVP | 1 (25) | Y181C |
| PI⁵, Total | 15 (100) | |
| APV | 1 (6.7) | L50V |
| IDV | 5 (33.3) | M46I |
| IDV, RTV | 1 (6.7) | V82A |
| NFV | 2 (13.3) | N88D |
| NFV, SQV | 6 (40) | L90M |
| MDR⁶, Total | 6 (100) | |
| AZT, d4T, ddC, ddi, ABC, DLV, EFV, NVP | 2 (33.3) | Q151M, K103N |
| AZT, d4T, DLV, EFV, NVP | 1 (16.7) | M41L, T215Y, K103N, Y181C |
| 3TC, DLV, EFV, NVP | 2 (33.3) | M184V, K103N |
| 3TC, NFV, SQV | 1 (16.7) | M184V, L90M |

¹ Major mutations were identified by sequencing the entire protease enzyme and the first 253 amino acids of reverse transcriptase.

² NRTI refers to nucleoside reverse transcriptase inhibitor. Other commonly used names for the NRTIs mentioned include AZT (zidovudine, retrovir); d4T (stavudine, zerit); ddC (zalcitabine, hivid); 3TC (lamivudine, epivir); and ABC, abacavir (1592, ziagen).

³ M41L refers to the substitution of amino acid methionine (M) by leucine (L) at position 41 of the reverse transcriptase enzyme. Other mutation nomenclature refers to substitutions as indicated; amino acid abbreviations are as follows: K, lysine; R, arginine; T, threonine; D, aspartic acid; N, asparagine; E, glutamic acid; H, histidine; Y, tyrosine; V, valine; I, isoleucine; A, alanine.

⁴ NNRTI refers to non-nucleoside reverse transcriptase inhibitor. Other commonly used names for the NNRTIs mentioned include DLV, delavirdine (rescriptor); EFV, efavirenz (sustiva); and NVP, nevirapine (viramune).

⁵ PI refers to protease inhibitor. Other commonly used names for the PIs mentioned include SQV, saquinavir (invirase, fortovase); RTV, ritonavir (norvir); APV, amprenavir (agenerase); NFV, nelfinavir (viracept); IDV, indinavir (crixivan)

⁶ MDR refers to multi-drug resistance and includes mutations in HIV-1 that are associated with resistance to any two of the three classes of antiretroviral drugs (NRTIs, NNRTIs and protease inhibitors).

Appendix 2 contains a list of the major mutations associated with drug resistance that were used in the generation of this report. The major mutations that have been added to our consensus list since the last surveillance report of samples received to June 30, 2001, include I50L, D67N, Y115F, L210W, K219E/Q, L100I, V108I, Y188C/H/L, P225H, M230L and P236L. Table 2 shows the major mutations in the reverse transcriptase and protease genes of HIV-1 that are associated with resistance to NRTIs, NNRTIs, and protease inhibitors. Of the 35 individuals harbouring HIV-1 with mutations associated with resistance to NRTIs, the majority (21, 60%) carried virus with an M41L mutation in reverse transcriptase. The mutation M41L refers to the replacement of the amino acid methionine (M) with leucine (L) at position 41 of the reverse transcriptase enzyme. The majority (28, 80%) harboured HIV-1 resistant to AZT and d4T. Other mutations were also identified, which were associated with resistance to ddC, 3TC and adefovir. A total of four individuals harboured virus resistant to NNRTIs. Major mutations associated with resistance to all currently approved NNRTIs (delavirdine, efavirenz, and nevirapine) were identified in the sample population. Of 15 individuals harbouring major mutations associated with protease resistance, the majority (5, 33.3%) carried virus with an M46I mutation associated with resistance to indinavir; M46I refers to the replacement of methionine with isoleucine at position 46 in the protease enzyme. Major mutations associated with resistance to the protease inhibitors amprenavir, ritonavir, nelfinavir, and saquinavir were also identified in the sample population.

Table 3 shows the prevalence of primary drug resistance in the sample population by year of diagnosis with HIV infection. Larger and more representative samples are required to conduct trend analyses and determine significant associations with primary drug resistance. However, the following observations can be cautiously made: resistance to NRTIs has been observed in treatment naïve people with newly diagnosed infection as early as 1998; resistance to protease inhibitors and multi-drug resistance in this population has been observed as early as 1999; resistance to NRTIs and protease inhibitors may have reached a plateau since 1999/2000; and resistance to NNRTI and multi-drug resistance may be increasing over time in the sample population.

Table 3: Prevalence of drug resistance among treatment naïve individuals with newly diagnosed infection by year of diagnosis

| | Primary drug resistance | | | | | Total |
|-------------------|----------------------------------------|-------------------|--------------------|-----------------------|------------------|------------------|
| | Wild type/minor mutations ¹ | NRTI ² | NNRTI ³ | Protease ⁴ | MDR ⁵ | |
| Year of diagnosis | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) |
| 1997 | 20 (100) | 0 | 0 | 0 | 0 | 20 (100) |
| 1998 | 46 (90.2) | 5 (9.8) | 0 | 0 | 0 | 51 (100) |
| 1999 | 250 (92.3) | 14 (5.1) | 0 | 6 (2.2) | 1 (0.4) | 271 (100) |
| 2000 | 279 (95.9) | 7 (2.4) | 1 (0.3) | 4 (1.4) | 0 | 291 (100) |
| 2001 | 155 (89.1) | 9 (5.2) | 3 (1.7) | 4 (2.3) | 3 (1.7) | 174 (100) |
| Total | 750 (92.9) | 35 (4.3) | 4 (0.5) | 14 (1.8) | 4 (0.5) | 807 (100) |

¹ Wild type indicates that no major mutations associated with drug resistance were identified. Minor mutations refers to genetic variables not associated with drug resistance. Year of diagnosis was unknown for 37 individuals infected with wild type virus or HIV-1 with minor mutation.

² NRTI refers to nucleoside reverse transcriptase inhibitor. Year of diagnosis was unknown for one individual infected with HIV-1 harbouring a major mutation to an NRTI.

³ NNRTI refers to non-nucleoside reverse transcriptase inhibitor.

⁴ Year of diagnosis was unknown for one individual harbouring major mutations associated with resistance to a protease inhibitor.

⁵ MDR refers to multi-drug resistance and includes mutations in HIV-1 that are associated with resistance to any two of the three classes of antiretroviral drugs (NRTIs, NNRTIs and protease inhibitors). Year of diagnosis was unknown for two individuals harbouring multi-drug resistant HIV-1.

Table 4 shows the prevalence of primary drug resistance in the sample population by province. Larger samples that are more representative of all newly diagnosed cases will help determine significant associations with primary drug resistance. However, the following observations can be cautiously made: drug resistance has been identified in BC, Alberta and Manitoba; resistance to all three classes of approved antiretroviral drugs has been identified in BC; multi-drug resistance has been identified in treatment naïve individuals with newly diagnosed infection in BC and Alberta. Resistance to the protease inhibitors nelfinavir and saquinavir has been identified in one sample obtained from Ontario through the sentinel arm of the CHSDRSP.

Table 4: Prevalence of drug resistance among treatment naïve individuals with newly diagnosed infection by province

| | Primary drug resistance | | | | | Total |
|--------------------------|----------------------------------------|-------------------|--------------------|-----------------|------------------|------------------|
| | Wild type/minor mutations ¹ | NRTI ² | NNRTI ³ | Protease | MDR ⁴ | |
| Province | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) |
| British Columbia | 508 (94.6) | 16 (3.1) | 4 (0.7) | 5 (0.9) | 4 (0.7) | 537(100) |
| Alberta | 181 (92.8) | 7 (3.6) | 0 | 5 (2.6) | 2 (1) | 195 (100) |
| Saskatchewan | 33 (100) | 0 | 0 | 0 | 0 | 33 (100) |
| Manitoba | 64 (79) | 12 (14.8) | 0 | 5 (6.2) | 0 | 81 (100) |
| Nova Scotia ⁵ | 1 (100) | 0 | 0 | 0 | 0 | 1 (100) |
| Total | 787 (92.9) | 35 (4.1) | 4 (0.5) | 15 (1.8) | 6 (0.7) | 847 (100) |

¹ Wild type indicates that no major mutations associated with drug resistance were identified. Minor mutations refers to genetic variables not associated with drug resistance.

² NRTI refers to nucleoside reverse transcriptase inhibitor.

³ NNRTI refers to non-nucleoside reverse transcriptase inhibitor.

⁴ MDR refers to multi-drug resistance and includes mutations in HIV-1 that are associated with resistance to any two of the three classes of antiretroviral drugs (NRTIs, NNRTIs and protease inhibitors).

⁵ Amplification was successful for one out of nine samples received from individuals with newly diagnosed infection during 2001 in Nova Scotia.

In Tables 5 to 10, NNRTIs and NRTIs have been grouped together as reverse transcriptase inhibitors because of the small samples and in order to ensure the confidentiality of affected individuals.

Tables 5 and 6 show primary drug resistance in the sample population by age and gender. While the data are not representative of all newly diagnosed cases between 1997 and 2001, they demonstrate that primary drug resistance has been identified in adults of both genders between the ages of 15 and 80 years at first diagnosis of HIV infection.

Table 5: Prevalence of drug resistance among treatment naïve individuals with newly diagnosed HIV infection by age at diagnosis

| | Primary drug resistance | | | | |
|--------------------------|----------------------------------------|------------------|-----------------------|------------------|------------------|
| | Wild type/minor mutations ¹ | RTI ² | Protease ³ | MDR ⁴ | Total |
| Age at diagnosis (years) | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) |
| < 15 | 3 (100) | 0 | 0 | 0 | 3 (100) |
| 15-19 | 9 (90) | 0 | 1 (10) | 0 | 10 (100) |
| 20-29 | 134 (90.5) | 9 (6.1) | 3 (2) | 2 (1.4) | 148 (100) |
| 30-39 | 254 (92) | 16 (5.8) | 6 (2.2) | 0 | 276 (100) |
| 40-49 | 194 (96) | 5 (2.5) | 6 (1.5) | 0 | 202 (100) |
| 50-59 | 54 (98.2) | 1 (1.8) | 0 | 0 | 55 (100) |
| ≥ 60 | 26 (86.7) | 3 (10) | 1 (3.3) | 0 | 30 (100) |
| Total | 674 (93.1) | 34 (4.7) | 14 (1.9) | 2 (0.3) | 724 (100) |

¹ Wild type indicates that no major mutations associated with drug resistance were identified. Minor mutations refers to genetic variables not associated with drug resistance. Age at diagnosis was unknown for 113 individuals infected with wild type virus or HIV-1 harbouring minor mutation.

² RTI refers to reverse transcriptase inhibitor and includes both nucleoside and non-nucleoside reverse transcriptase inhibitors. Age at diagnosis was unknown for five individuals harbouring HIV-1 with mutations associated with resistance to RTIs.

³ Age at diagnosis was unknown for one individual harbouring HIV-1 with mutations associated with protease inhibitors.

⁴ MDR refers to multi-drug resistance and includes mutations in HIV-1 that are associated with resistance to any two of the three classes of antiretroviral drugs (NRTIs, NNRTIs, and protease inhibitors). Age at diagnosis was unknown for four individuals harbouring multi-drug resistant HIV-1.

Table 6: Prevalence of drug resistance among treatment naïve individuals with newly diagnosed infection by gender

| | Primary drug resistance | | | | |
|--------------|----------------------------------------|------------------|-----------------------|------------------|------------------|
| | Wild type/minor mutations ¹ | RTI ² | Protease ³ | MDR ⁴ | Total |
| Gender | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) |
| Male | 528 (93.1) | 30 (5.3) | 8 (1.4) | 1 (0.2) | 567 (100) |
| Female | 156 (92.3) | 6 (3.6) | 6 (3.6) | 1 (0.5) | 169 (100) |
| Total | 684 (92.9) | 36 (4.9) | 14 (1.9) | 2 (0.3) | 736 (100) |

¹ Wild type indicates that no major mutations associated with drug resistance were identified. Minor mutations refers to genetic variables not associated with drug resistance. Gender was unknown for 103 individuals infected with wild type virus or HIV-1 harbouring minor mutation.

² RTI refers to reverse transcriptase inhibitor and includes both nucleoside and non-nucleoside reverse transcriptase inhibitors. Gender was unknown for three individuals harbouring HIV-1 with mutations associated with resistance to RTIs.

³ Gender was unknown for one individual harbouring HIV-1 with mutations associated with protease inhibitors.

⁴ MDR refers to multi-drug resistance and includes mutations in HIV-1 that are associated with resistance to any two of the three classes of antiretroviral drugs (NRTIs, NNRTIs, and protease inhibitors).

Table 7 shows the prevalence of primary drug resistance by exposure category. The data are not representative of all newly diagnosed cases of HIV between 1997 and 2001, and because of the large proportion of individuals with unknown risk factors and the small samples in certain cells, significant associations between risk exposure and primary drug resistance could not be determined. However, the data indicate that primary drug resistance was found in the following risk exposures: male to male sex, injecting drug use, and heterosexual contact, specifically heterosexual contact with a person at risk of HIV infection.

Table 7: Prevalence of drug resistance among treatment naïve individuals with newly diagnosed infection by exposure category

| Exposure category | Primary drug resistance | | | | |
|---------------------------------------|----------------------------------------|------------------|-----------------|------------------|------------------|
| | Wild type/minor mutations ¹ | RTI ² | Protease | MDR ³ | Total |
| | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) |
| MSM ⁴ | 187 (92.1) | 14 (6.9) | 1 (0.5) | 1 (0.5) | 203 (100) |
| MSM/IDU ⁵ | 21 (84) | 3 (12) | 1 (4) | 0 | 25 (100) |
| IDU | 215 (95.6) | 5 (2.2) | 5 (2.2) | 0 | 225 (100) |
| Blood/blood products | | | | | |
| a) recipient of blood | 3 (100) | 0 | 0 | 0 | 3 (100) |
| b) recipient of clotting factor | 0 | 0 | 0 | 0 | 0 |
| Heterosexual contact/endemic | | | | | |
| a) origin in pattern II country | 11 (100) | 0 | 0 | 0 | 11 (100) |
| b) sexual contact with person at risk | 65 (92.9) | 4 (5.7) | 1 (1.4) | 0 | 70 (100) |
| Occupational exposure | 0 | 0 | 0 | 0 | 0 |
| NIR-HET ⁶ | 85 (87.6) | 8 (8.3) | 4 (4.1) | 0 | 97 (100) |
| Other | 0 | 0 | 0 | 0 | 0 |
| NIR ⁷ | 199 (93.8) | 5 (2.4) | 3 (1.4) | 5 (2.4) | 212 (100) |
| Perinatal | 1 (100) | 0 | 0 | 0 | 1 (100) |
| Total | 787 (92.9) | 39 (4.6) | 15 (1.8) | 6 (0.7) | 847 (100) |

¹ Wild type indicates that no major mutations associated with drug resistance were identified. Minor mutations refers to genetic variables not associated with drug resistance.

² RTI refers to reverse transcriptase inhibitor and includes both nucleoside and non-nucleoside reverse transcriptase inhibitors.

³ MDR refers to multi-drug resistance and includes mutations in HIV-1 that are associated with resistance to any two of the three classes of antiretroviral drugs (NRTIs, NNRTIs, and protease inhibitors).

⁴ MSM refers to men who have sex with men.

⁵ IDU refers to injecting drug use.

⁶ NIR-HET refers to non-identified risk related to heterosexual exposure.

⁷ NIR refers to non-identified risk exposures, i.e. when exposures are not identified.

Table 8 shows the prevalence of primary drug resistance by ethnicity. The data are not representative of all newly diagnosed cases of HIV between 1997 and 2001, and because of small samples in certain cells significant associations between ethnicity and primary drug resistance could not be determined. However, the data suggest that while the majority of primary drug resistance (34 cases, 75.6%) was identified in White individuals in the sample population, primary drug resistance has also been identified in affected Aboriginal and Asian people.

Table 8: Prevalence of drug resistance among treatment naïve individuals with newly diagnosed infection by ethnicity

| | Primary drug resistance | | | | |
|------------------|----------------------------------------|------------------|-----------------------|------------------|------------------|
| | Wild type/minor mutations ¹ | RTI ² | Protease ³ | MDR ⁴ | Total |
| Ethnicity | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) |
| White | 402 (92.2) | 22 (5) | 10 (2.3) | 2 (0.5) | 436 (100) |
| Black | 35 (97.2) | 1 (2.8) | 0 | 0 | 36 (100) |
| Aboriginal | | | | | |
| Native Indian | 68 (94.4) | 2 (2.8) | 2 (2.8) | 0 | 72 (100) |
| Metis | 13 (100) | 0 | 0 | 0 | 13 (100) |
| Inuit | 3 (100) | 0 | 0 | 0 | 3 (100) |
| Unspecified | 37 (90.2) | 3 (7.3) | 1 (2.4) | 0 | 41 (100) |
| Asian | 12 (92.3) | 1 (7.7) | 0 | 0 | 13 (100) |
| South Asian | 6 (85.7) | 1 (14.3) | 0 | 0 | 7 (100) |
| Latin-American | 11 (100) | 0 | 0 | 0 | 11 (100) |
| Other (mixed) | 20 (100) | 0 | 0 | 0 | 20 (100) |
| Total | 607 (93.1) | 30 (4.6) | 13 (2) | 2 (0.3) | 652 (100) |

¹ Wild type indicates that no major mutations associated with drug resistance were identified. Minor mutations refers to genetic variables not associated with drug resistance. Ethnicity was unknown for 180 individuals infected with wild type virus or HIV-1 with minor mutations.

² RTI refers to reverse transcriptase inhibitor and includes both nucleoside and non-nucleoside reverse transcriptase inhibitors. Ethnicity was unknown for nine individuals infected with HIV-1 harbouring mutations associated with RTIs.

³ Ethnicity was unknown for two individuals infected with HIV-1 harbouring mutations associated with protease inhibitors.

⁴ MDR refers to multi-drug resistance and includes mutations in HIV-1 that are associated with resistance to any two of the three classes of antiretroviral drugs (NRTIs, NNRTIs, and protease inhibitors). Ethnicity was unknown for four individuals harbouring multi-drug resistant HIV-1.

Table 9 shows the prevalence of primary drug resistance by HIV-1 subtype. The data are not representative of all newly diagnosed cases of HIV between 1997 and 2001, and because of small samples in certain cells significant associations between HIV-1 subtype and primary drug resistance could not be determined. However, the data suggest that while the majority (56 cases, 96.6%) of primary drug resistance has been identified in individuals with HIV-1 subtype B, primary drug resistance has also been identified in individuals infected with HIV-1 subtypes C and A. Resistance to the protease inhibitors nelfinavir and saquinavir was identified in one HIV-1 subtype C sample from Ontario that was received through the sentinel arm of the CHSDRSP.

Table 9: Prevalence of drug resistance among treatment naïve individuals by HIV-1 subtype

| | Primary drug resistance | | | | Total |
|----------------|----------------------------------------|------------------|-----------------------|------------------|------------------|
| | Wild type/minor mutations ¹ | RTI ² | Protease ³ | MDR ⁴ | |
| HIV-1 subtype | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) |
| B | 729 (92.9) | 37 (4.7) | 13 (1.7) | 6 (0.7) | 785 (100) |
| A | 6 (85.7) | 0 | 1 (14.3) | 0 | 7 (100) |
| C | 29 (96.7) | 1 (3.3) | 0 | 0 | 30 (100) |
| D | 3 (100) | 0 | 0 | 0 | 3 (100) |
| E ⁵ | 3 (100) | 0 | 0 | 0 | 3 (100) |
| A/C | 1 (100) | 0 | 0 | 0 | 1 (100) |
| A/G | 1 (100) | 0 | 0 | 0 | 1 (100) |
| Total | 772 (93) | 38 (4.6) | 14 (1.7) | 6 (0.7) | 830 (100) |

¹ Wild type indicates that no major mutations associated with drug resistance were identified. Minor mutations refers to genetic variables not associated with drug resistance. HIV-1 subtype was unknown for 15 individuals infected with wild type virus or HIV-1 with minor mutations.

² RTI refers to reverse transcriptase inhibitor and includes both nucleoside and non-nucleoside reverse transcriptase inhibitors. Subtype was unknown for one individual infected with HIV-1 harbouring mutations associated with RTIs.

³ HIV-1 subtype was unknown for one individual infected with HIV-1 harbouring mutations associated with protease inhibitors.

⁴ MDR refers to multi-drug resistance and includes mutations in HIV-1 that are associated with resistance to any two of the three classes of antiretroviral drugs (NRTIs, NNRTIs, and protease inhibitors).

Table 10 shows the prevalence of primary drug resistance among recently acquired (within approximately the previous 4 months) versus prevalent infections. The lack of availability of test kits aimed at determining incident infections has affected the extent to which these data could be generated. The sample size is therefore not reflective of all newly diagnosed cases between 1997 and 2001 or samples for which drug resistance genotyping has been completed. Therefore, significant associations between time of infection and primary drug resistance could not be determined. However, the data suggest that primary drug resistance has been identified in both prevalent and recent infections. These data also suggest that certain mutations may persist over time and contribute to drug resistance.

Table 10: Prevalence of drug resistance among treatment naïve individuals with recently acquired versus prevalent HIV-1 infection

| | Primary drug resistance | | | | Total |
|---------------------|----------------------------------------|------------------|----------------|------------------|------------------|
| | Wild type/minor mutations ¹ | RTI ² | Protease | MDR ³ | |
| HIV-1 Infection | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) |
| Recent ⁴ | 144 (90.6) | 9 (5.7) | 4 (2.5) | 2 (1.2) | 159 (100) |
| Prevalent | 378 (95.5) | 15 (3.8) | 3 (0.7) | 0 | 396 (100) |
| Total | 522 (94.1) | 24 (4.3) | 7 (1.3) | 2 (0.3) | 555 (100) |

¹ Wild type indicates that no major mutations associated with drug resistance were identified. Minor mutations refers to genetic variables not associated with drug resistance. Out of 524 individuals infected with wild type virus or HIV-1 harbouring minor mutations, time of HIV-1 infection was unknown for two individuals.

² RTI refers to reverse transcriptase inhibitor and includes both nucleoside and non-nucleoside reverse transcriptase inhibitors.

³ MDR refers to multi-drug resistance and includes mutations in HIV-1 that are associated with resistance to any two of the three classes of antiretroviral drugs (NRTIs, NNRTIs, and protease inhibitors).

⁴ Because of kit availability, a combination of three assays (Organon Technika, Avidity Index, and Abbott) was used to determine recent infection. If any one test identified a sample as representing recent infection, the sample was included in this category.

Table 11 is a summary of primary drug resistance results from the CHSDRSP and other cohort and cross-sectional studies in Canada. Please note that this table is NOT meant for inter-study comparisons. It is difficult to make such comparisons and arrive at firm conclusions because of differences in study design. For example, prevalence rates depend on the population being studied (high risk versus general population), the types of laboratory tests used (genotypic and/or phenotypic testing) and differences in the mutations studied and reported. The results suggest that, in Canada, the overall prevalence of primary drug resistance to RTIs is between 4.6% and 20.0%; primary drug resistance to protease inhibitors is between 1.4% and 6.0%. Primary drug resistance to more than one class of antiretroviral drug (multi-drug resistance) has been observed in Canada, and preliminary studies suggest an overall prevalence of between 0.4% and 9.9%.

Table 11: Summary of key studies on drug resistance among treatment naïve individuals with newly diagnosed infection in Canada

| Province ¹ | Year of diagnosis | Risk exposures ² | Sample size | RTI ³ % | Protease ⁴ % | MDR ⁵ % | Total % |
|----------------------------------|-----------------------------------------------|-----------------------------|-------------|-----------------------|----------------------------|-----------------------|-------------|
| BC ⁶ | 1997-1998 | Mixed | 423 | 4.6 (n = 416) | 4.6 | – | – |
| QC ⁷ | 1997-1999 | IDU (26%) Sexual (69%) | 81 | 20 | 6 | 9.9 | – |
| QC ⁸ | May 1996-June 2000 July 2000-December 2001 | Mixed | 112 | – | – | 4.1 | 23.2 |
| | | Mixed | 36 | – | – | 0 | 11.4 |
| ON ⁹ | 1997-1999 | MSM | 23 | 13 | – | – | – |
| BC, AB, SK, MB, NS ¹⁰ | 1997 | Mixed | 20 | 0 | 0 | 0 | 0 |
| | 1998 | Mixed | | 9.8 (NRTI) | 0 | 0 | 9.8 |
| | 1999 | Mixed | 51 | 5.1 (NRTI) | 2.2 | 0.4 | 7.7 |
| | 2000 | Mixed | 271 | 2.4 (NRTI) | 1.4 | 0 | 4.1 |
| | 2001 | Mixed | 291 | 0.3 (NNRTI) | | | |
| | | | 174 | 5.2 (NRTI) | 2.3 | 1.7 | 10.9 |
| | | | | 1.7 (NNRTI) | | | |

¹ Province abbreviations: British Columbia (BC); Quebec (QC); Ontario (ON); Alberta (AB); Saskatchewan (SK); Manitoba (MB); Nova Scotia (NS).

² IDU refers to injecting drug use.

³ RTI refers to reverse transcriptase inhibitors. Differentiation into nucleoside and non-nucleoside reverse transcriptase inhibitors (NRTI and NNRTI respectively) is given where this information was available. Only major mutations associated with RTI resistance are identified.

⁴ Only major mutations associated with protease resistance are identified.

⁵ MDR refers to multi-drug resistance and includes mutations in HIV-1 that are associated with resistance to any two of the three classes of antiretroviral drugs (NRTIs, NNRTIs and PIs).

⁶ Alexander CS et al. 8th Annual Canadian Conference on HIV/AIDS Research, Vancouver, BC, May 1999: #B224.

⁷ Saloman H et al. AIDS 2000; 142(2):F17-23.

⁸ Routy JP et al. XI International Drug Resistance Workshop, Seville, Spain, July 2002: Antiviral Ther 7 (Suppl. 1): #179.

⁹ Cassol S et al. 9th Annual Canadian Conference on HIV/AIDS Research, Montreal, QC, April 2000 #135P.

¹⁰ The Canadian HIV Strain and Drug Resistance Surveillance Program (this report).

Table 12 shows the results from studies on primary drug resistance that were conducted in the United States and other countries in western Europe. Please note that this table is NOT meant for inter-study comparisons; such interpretations are difficult to make because of the differences in study design. The results suggest that the prevalence of major mutations associated with drug resistance is similar to that described in Canada.

Table 12. Summary of key studies on drug resistance among treatment naïve individuals with newly diagnosed infection in the United States and Western Europe

| Country | Year of diagnosis | Risk exposure ¹ | Sample size | RTI ² % | Protease ³ % | MDR ⁴ % | Total ⁵ % |
|------------------------------------------------------|-------------------|----------------------------|-------------|---------------------------------------------|-------------------------|--------------------|----------------------|
| United States ⁶ | 1989-1998 | MSM (80%) | 141 | 0.7 (NNRTI) | 1.4 | 1.4 | 2.1 |
| United States ⁷ | 1995-1999 | MSM (94%) | 80 | 12.5 (NRTI) 7.5 (NNRTI) | 3.0 | 3.8 | 16.3 |
| United States ⁸ | 1997-1998 | – | 144 | 4.0 (NRTI) 15.0 (NNRTI, n = 95) | 10.0 | 5.0 | 22.0 |
| United States ⁹ | 1998 | Mixed | 238 | 3.4 (NRTI) 0.4 (NNRTI) | 0 | 0 | 3.8 |
| | 1999 | | 240 | 8.3 (NRTI) 2.1 (NNRTI) | 1.7 | 1.7 | 10.0 |
| | 2000 | | 245 | 6.9 (NRTI) 1.2 (NNRTI) | 2 | 1.2 | 9.0 |
| United States (Montreal and Vancouver) ¹⁰ | 1995-1998 | MSM | 377 | 8.5 (NRTI, n = 213) 1.7 (NNRTI, n = 176) | 0.9 (n = 213) | 3.8 (n = 213) | 8.0 |
| | 1999-2000 | | | 15.9 (NNRTI, n = 82) 7.3 (NNRTI, n = 82) | 9.1 (n = 88) | 10.2 (n = 88) | 22.7 |
| France ¹¹ | 1995-1998 | Mixed | 48 | 16.6 | 2.0 | – | – |
| France ¹² | 1999-2000 | Mixed | 251 | 7.6 (NRTI) 4.0 (NNRTI) | 5.2 | 4.8 | – |
| Spain ¹³ | 1996-1998 | Mixed | 68 | 16.2 | 6 | 4.4 | – |
| Spain ¹⁴ | 1997-1999 | Mixed | 31 | 16.1 | 9.7 | 0 | 25.8 |
| | 2000-2001 | | 21 | 0 | 4.8 | 0 | 4.8 |
| Switzerland ¹⁵ | 1996 | Mixed | 193 | 5.6 | 3 | – | 8.6 |
| | 1997 | | | 6.9 | 7.7 | – | 14.6 |
| | 1998 | | | 6.8 | 2.0 | – | 8.8 |
| | 1999 | | | 3.1 | 1.9 | – | 5.0 |
| Switzerland ¹⁶ | 1999-2001 | Mixed | 200 | 6.5 (NRTI) 0.5 (NNRTI) | 1.0 | 1.5 | 10 |
| United Kingdom ¹⁷ | 1994-1996 | Mixed | 21 | 0 | 0 | – | 0 |
| | 1997-1999 | Mixed | 22 | 13.6 | 0 | 0 | 13.6 |
| | 2000 | Mixed | 26 | 19.2 | 3.8 | 0 | 23.0 |

¹ MSM refers to men who have sex with men.

² RTI refers to reverse transcriptase inhibitors. NRTI = nucleoside reverse transcriptase inhibitor, NNRTI = Non-nucleoside reverse transcriptase inhibitor. Information on NRTI and NNRTI provided when available.

³ Protease refers to protease inhibitors.

⁴ MDR refers to multi-drug resistance.

⁵ Total may include major and minor mutations associated with primary drug resistance.

⁶ Little SJ et al. JAMA 1999; 282:1142-49.

⁷ Boden D et al. JAMA 1999; 282:1135-41.

⁸ Wegner S et al. AIDS 2000; 14:1009-15.

⁹ Bennet D et al. 9th Conference on Retroviruses and Opportunistic Infections. Seattle WA. Feb 2002: #372M.

¹⁰ Little SJ et al. N Engl J Med 2002; 347(6):385-94.

¹¹ Tamalet C et al. J Med Virol 2000;61:181-86.

¹² Chaix ML et al. XI International HIV Drug Resistance Workshop. Seville, Spain. July 2002; Antiviral Ther 7(Suppl 1): #166.

¹³ Puig T et al. AIDS 2000; 14:727-32.

¹⁴ De Mendoza C, del Romero J, Rodriguez C, et al. 9th Conference on Retroviruses and Opportunistic Infections. Seattle WA. Feb 2002:371M.

¹⁵ Yerly S et al. AIDS 2001; 15:2287-92.

¹⁶ Yerly S et al. XI International HIV Drug Resistance Workshop. Seville, Spain. July 2002; Antiviral Ther 7 (Suppl 1): #183.

¹⁷ UK Collaborative Group on Monitoring the Transmission of HIV Drug Resistance. BMJ 2001; 322:1087-88.

4.0 HIV-1 Subtypes (1984-January 2002)

4.1 Background

Since the first reported cases of HIV/AIDS in the mid-1980s, HIV has emerged as one of the most significant infectious agents, infecting 40 million people worldwide. Key to the pathogenicity of this virus is its genetic heterogeneity, which is the result of the error-prone reverse transcriptase, the rapid turnover of HIV-1 *in vivo*, recombination, and selective immune pressures by the host.

The initial classification of HIV into two main types, HIV-1 and HIV-2, was based on the geographic distribution and the animal source of the human infection – chimpanzee (*Pan troglodytes*) for HIV-1 and sooty mangabey (*Cercocebus atys*) for HIV-2. Expanding access to diverse samples of HIV-1 and the advent of new molecular tools have led to the classification of HIV-1 into three distantly related “groups”: M (for main), N (for non-M, non-O) and O (for outlier). Distinct lineages within group M have also been identified. These include subtype designations A to E (subtype E is also referred to as CRF01_AE [the circulating recombinant form, CRF A/E]), F to H, J and K.

One of the primary public health related reasons for conducting a systematic surveillance of HIV genetic variability in Canada is to inform vaccine research and development. The majority of vaccines undergoing clinical trials have been developed using strains of HIV-1 subtype B (predominant in North America and Europe). However, the ability of these vaccines to elicit cross-subtype responses is unclear. It is therefore imperative to collect data on the prevalence and incidence of HIV subtypes and other determinants of subtype diversity in Canada in order to guide vaccine strategies.

Another reason for conducting the systematic surveillance of HIV genetic diversity is to determine whether currently approved assays for HIV in Canada are capable of detecting all circulating strains. This includes our ability to control and manage HIV infection through approved viral load test assays and other tests that determine the stage and progression of the disease. This topic is addressed in more detail in Section 5, National HIV Reference Services.

The data presented in this report deal with samples that were received by the National Laboratory for HIV Genetics as of June 30, 2002, and represent newly diagnosed cases of HIV-1 infection between 1984 and 2002.

4.2 Data sources

This section highlights the main findings from the CHSDRSP up to June 30, 2002. It is important to note that the results presented here represent individuals who sought testing, whose condition was properly diagnosed and reported as HIV positive. Furthermore, the results include only those individuals for whom sufficient sera, taken for the purposes of diagnostic testing, were available to send to the national HIV laboratories by June 30, 2002, and, of these samples, the subset for which RT-PCR amplification and sequencing to identify major mutations were successful.

As of June 30, 2002, serum samples from 2,242 individuals with HIV infection newly diagnosed between 1984 and 2002 and corresponding non-nominal epidemiologic data have been received from British Columbia (BC), Alberta, Manitoba, Saskatchewan, Newfoundland and Labrador and Nova Scotia for HIV-1 subtype analysis. A total of 32 serum samples have also been received from Ontario, but since these samples were received through the sentinel arm of the CHSDRSP the results are reported in Section 5 under National HIV Reference Services. While the goal of the CHSDRSP is to collect serum samples from all newly diagnosed cases, the data presented in this report are a result of convenience sampling methods and may not be representative. As well, discussions are under way to expand this program to the remaining provinces and territories.

At the time of writing this report (December 2002) the National Laboratory for HIV Genetics had analyzed a total of 1,634 samples for HIV-1 subtype. Viral RNA had been successfully amplified from 1,312 (80.3%) of the serum samples. This level of success in amplifying virus from serum specimens will likely improve further as sample quality is enhanced and through the identification and use of various primer combinations for reverse transcriptase PCR amplification.

4.3 Prevalence and determinants of HIV-1 subtypes in sample population (N = 1,312)

Table 13 provides information on the distribution of HIV-1 subtypes in our sample population. Between July 1998 and December 2000, the C2-V5 region (233 amino acids) of the envelope protein was used to assess HIV subtype. Since this time, sequence analysis of the *pol* gene (entire protease and the first 253 amino acids of reverse transcriptase) has been used for subtype analysis. While the majority (93.1%) of samples are of HIV-1 subtype B, other subtypes have been identified. In decreasing order of prevalence they include subtype C (4.1%), A (1.8%), E (0.4%), D (0.3%) and the recombinants A/B (0.1%), A/C (0.1%) and A/G (0.1%). The recombinant A/G, from two individuals, has been identified in Ontario from samples sent through the sentinel arm of the CHSDRSP. See Section 5, National HIV Reference Services, for further information.

Table 13: Prevalence of HIV-1 subtypes among treatment naïve individuals with newly diagnosed infection (N = 1,312)

| HIV-1 subtype | Frequency | Percentage |
|----------------|--------------|------------|
| A | 24 | 1.8 |
| A/B | 1 | 0.1 |
| A/C | 1 | 0.1 |
| A/G | 1 | 0.1 |
| B | 1,222 | 93.1 |
| C | 54 | 4.1 |
| D | 4 | 0.3 |
| E ¹ | 5 | 0.4 |
| Total | 1,312 | 100 |

¹ HIV-1 subtype E has also been referred to as the circulating recombinant form (CRF) A/E.

Although existing studies on high-risk populations also suggest the predominance of HIV-1 subtype B in Canada, subtype A was reported in Canada in 1995^{*}. As of November 2000, all samples of 31 recent seroconverters from the POLARIS cohort (comprising men who have sex with men) in Ontario are of subtype B^{**}. The British Columbia Centre for Excellence in HIV/AIDS has identified subtypes A, C and D in at least 4% of individuals linked to cohort studies and to the BC HIV Drug Treatment Program^{***}. All HIV-1 sequences analyzed from injecting drug users ($n = 17$) and men who have sex with men ($n = 5$) residing in Montreal were of subtype B^{****}.

Table 14 shows the prevalence of HIV-1 subtypes by year of diagnosis with HIV infection. The results suggest a decrease in the prevalence of non-B HIV-1 subtypes, from 22.7% during 1996 to 1.7% during 2001. However, the majority of samples from 1996 were from BC, and the samples are not representative of newly diagnosed cases of HIV infection in the noted year. Therefore, significant associations between year of first diagnosis and HIV-1 subtype could not be determined.

* Montpetit M. HIV-1 Subtype A in Canada. *AIDS Res Hum Retroviruses*. 1995;11(11):1421-22.

** Major C, for the POLARIS Seroconverter Study Group. Proceedings of the Division of HIV Epidemiology and Surveillance, Annual Meeting, BHST, CIDPC, Health Canada. Halifax, Nov. 16-18 2000.

*** Alexander C, Dong W, Chan K, et al. HIV-1 non-B Subtypes in a Large North American Cohort: Prevalence and Response to Antiretroviral Therapy. 7th Conference on Retroviruses and Opportunistic Infections. San Francisco, CA. Jan. 31-Feb 3, 2000; #174.

**** Bernier L, Lamothe F, Bruneau J, et al. Eighth Annual Canadian Conference on AIDS, Vancouver BC May 1-4 1999: #104A.

Table 14: Prevalence of HIV-1 subtypes by year of first diagnosis with HIV-1 infection

| | HIV-1 subtype | | | | | | | | |
|--------------------|-----------------|---------------------|-----------------|----------------|----------------|-----------------|-----------------|-----------------|--------------------|
| | A ¹ | B ² | C ³ | D | E ⁴ | A/B | A/C | A/G | Total |
| Year of diagnosis | n (%) | n (%) | n (%) | n (%) | n (%) | n (%) | n (%) | n (%) | n (%) |
| 1995 and before | 3 (4.5) | 63 (94) | 1 (1.5) | 0 | 0 | 0 | 0 | 0 | 67 (100) |
| 1996 | 3 (4.5) | 51 (77.3) | 11 (16.7) | 0 | 0 | 1 (1.5) | 0 | 0 | 66 (100) |
| 1997 | 6 (5.7) | 97 (92.3) | 1 (1) | 0 | 1 (1) | 0 | 0 | 0 | 105 (100) |
| 1998 | 2 (1.3) | 141 (92.7) | 8 (5.3) | 1 (0.7) | 0 | 0 | 0 | 0 | 152 (100) |
| 1999 | 6 (1.8) | 311 (90.9) | 20 (5.8) | 2 (0.6) | 3 (0.9) | 0 | 0 | 0 | 342 (100) |
| 2000 | 2 (0.7) | 292 (95.2) | 9 (2.9) | 1 (0.3) | 1 (0.3) | 0 | 1 (0.3) | 1 (0.3) | 307 (100) |
| 2001 | 0 | 171 (98.3) | 3 (1.7) | 0 | 0 | 0 | 0 | 0 | 174 (100) |
| January 2002 | 0 | 4 (10) | 0 | 0 | 0 | 0 | 0 | 0 | 4 (100) |
| Total n (%) | 22 (1.8) | 1,130 (92.9) | 53 (4.4) | 4 (0.3) | 5 (0.4) | 1 (0.08) | 1 (0.08) | 1 (0.08) | 1,217 (100) |

¹ Year of diagnosis was unknown for two individuals with HIV-1 subtype A infection.

² Year of diagnosis was unknown for 92 individuals with HIV-1 subtype B infection.

³ Year of diagnosis was unknown for one individual with HIV-1 subtype C infection.

⁴ HIV-1 subtype E has also been referred to as the circulating recombinant form (CRF) A/E.

Table 15 shows the prevalence of HIV-1 subtypes by province of diagnosis. The data indicate geographic variation in the distribution of non-B HIV-1 subtypes. Whereas all 42 samples from Newfoundland and Labrador were identified as subtype B, 10.4%, 8.2%, 6.8% and 4.6% of the analyzed samples from Manitoba, Saskatchewan, BC and Alberta respectively belonged to non-B HIV-1 subtypes. BC had the greatest genetic variation in the non-B HIV-1 subtypes. It should be noted, however, that sample sizes are not representative of the total population with a diagnosis of HIV in each of the indicated provinces. Furthermore, the provinces of Quebec and Ontario, which report the highest prevalence of HIV infections, are not represented, so the results should be interpreted with caution. Subtypes C and A and the recombinant subtype A/G have been identified in samples submitted through the sentinel arm of the CHSDRSP, see Section 5, National HIV Reference Services, for further information.

Table 15: Prevalence of HIV-1 subtypes by province

| | HIV-1 subtype | | | | | | | | |
|---------------------------|-----------------|---------------------|-----------------|-----------------|----------------|-----------------|-----------------|-----------------|--------------------|
| | A | B | C | D | E ¹ | A/B | A/C | A/G | Total |
| Province | n (%) | n (%) | n (%) | n (%) | n (%) | n (%) | n (%) | n (%) | n (%) |
| B.C. | 10 (1.5) | 673 (93.2) | 29 (4) | 4 (0.6) | 3 (0.4) | 1 (0.1) | 1 (0.1) | 1 (0.1) | 722 (100) |
| Alberta | 0 | 207 (95.4) | 8 (3.7) | 0 | 2 (0.9) | 0 | 0 | 0 | 217 (100) |
| Saskatchewan | 6 (4.1) | 135 (91.8) | 6 (4.1) | 0 | 0 | 0 | 0 | 0 | 147 (100) |
| Manitoba | 8 (4.4) | 164 (89.6) | 11 (6) | 0 | 0 | 0 | 0 | 0 | 183 (100) |
| Nova Scotia ² | 0 | 1 (100) | 0 | 0 | 0 | 0 | 0 | 0 | 1 (100) |
| Newfoundland and Labrador | 0 | 42 (100) | 0 | 0 | 0 | 0 | 0 | 0 | 42 (100) |
| Total | 24 (1.8) | 1,222 (93.1) | 54 (4.1) | 4 (4.03) | 5 (0.4) | 1 (0.08) | 1 (0.08) | 1 (0.08) | 1,312 (100) |

¹ HIV-1 subtype E has also been referred to as the circulating recombinant form (CRF) A/E.

² Amplification was successful for one out of nine samples received from individuals with newly diagnosed infection during 2001 in Nova Scotia.

In Tables 16 to 21, HIV-1 subtypes D, E, A/B, A/C and A/G have been grouped together as "other" subtypes in order to ensure the confidentiality of affected individuals.

Table 16 shows the prevalence of HIV-1 subtypes by age of diagnosis. While the samples in certain age categories are small and the data are not representative of all newly diagnosed cases of HIV infection, the results identified non-B subtypes of HIV-1 in most age groups.

Table 16: Prevalence of HIV-1 subtypes by age of diagnosis with HIV-1 infection

| | HIV-1 subtype | | | | |
|--------------|-----------------|---------------------|-----------------|--------------------|--------------------|
| | A ¹ | B ² | C ³ | Other ⁴ | Total |
| Age (years) | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) |
| < 15 | 2 (33.3) | 3 (50) | 1 (6.7) | 0 | 6 (100) |
| 15-19 | 0 | 20 (100) | 0 | 0 | 20 (100) |
| 20-29 | 8 (3.1) | 233(90.3) | 14 (5.4) | 3 (1.2) | 258 (100) |
| 30-39 | 3 (0.7) | 413 (92.8) | 24 (5.4) | 5 (1.1) | 445 (100) |
| 40-49 | 6 (2.1) | 265 (93) | 10 (3.5) | 4 (1.4) | 285 (100) |
| 50-59 | 2 (2.3) | 82 (95.4) | 2 (2.3) | 0 | 86 (100) |
| ≥ 60 | 1 (2.6) | 35 (92.1) | 2 (5.3) | 0 | 38 (100) |
| Total | 22 (1.9) | 1,015 (92.3) | 53 (4.7) | 12 (1.1) | 1,138 (100) |

¹ Age at diagnosis was unknown for two individuals with HIV-1 subtype A infection.

² Age at diagnosis was unknown for 171 individuals with HIV-1 subtype B infection.

³ Age at diagnosis was unknown for one individual with HIV-1 subtype C infection.

⁴ Other includes HIV-1 subtypes D, E, A/B, A/C and A/G. HIV-1 subtype E has also been referred to as the circulating recombinant form (CRF) A/E.

Table 17 shows the prevalence of HIV-1 subtypes by gender. While the data are not representative of all newly diagnosed cases of HIV infection, the results indicate that the prevalence of non-B subtypes may be higher among females than among males (12.9% versus 5.5% respectively).

Table 17: Prevalence of HIV-1 subtypes by gender

| | HIV-1 subtype | | | | |
|--------------|-----------------|---------------------|-----------------|--------------------|--------------------|
| | A ¹ | B ² | C | Other ³ | Total |
| Sex | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) |
| Male | 13 (1.4) | 858 (94.5) | 29 (3.2) | 8 (0.9) | 908 (100) |
| Female | 9 (3.1) | 257 (87.1) | 25 (8.4) | 4 (1.4) | 295 (100) |
| Total | 22 (1.8) | 1,115 (92.7) | 54 (4.5) | 12 (1) | 1,203 (100) |

¹ Gender was unknown for two individuals with HIV-1 subtype A infection.

² Gender was unknown for 107 individuals with HIV-1 subtype B infection.

³ Other includes HIV-1 subtypes D, E, A/B, A/C, and A/G. HIV-1 subtype E has also been referred to as the circulating recombinant form (CRF) A/E.

Table 18 shows the prevalence of HIV-1 subtypes by exposure category. While the samples in certain risk exposure categories are small and the data are not representative of all newly diagnosed cases of HIV infection, the results suggest that a higher proportion of individuals infected through heterosexual contact (particularly with other individuals at risk of HIV infection or individuals from pattern II countries where non-B strains of HIV-1 prevail) may be harbouring non-B HIV-1 subtypes than individuals infected through male-to-male sex or through injecting drug use. One case of perinatally acquired HIV-1 subtype A has been identified. Receipt of blood or clotting factor was identified as the sole exposure category for seven individuals. Further investigation is required to determine the accuracy of this information.

Table 18: Prevalence of HIV-1 subtypes by exposure category

| Exposure Category | HIV-1 subtype | | | | Total <i>n (%)</i> |
|---------------------------------------|-------------------|---------------------|-------------------|------------------------------------|-----------------------|
| | A <i>n (%)</i> | B <i>n (%)</i> | C <i>n (%)</i> | Other ¹ <i>n (%)</i> | |
| MSM ² | 3 (0.9) | 326 (97) | 6 (1.8) | 1 (0.3) | 336 (100) |
| MSM/IDU ³ | 1 (2.3) | 42 (95.4) | 1 (2.3) | 0 | 44 (100) |
| IDU | 2 (0.6) | 349 (97.4) | 6 (1.7) | 1 (0.3) | 358 (100) |
| Blood/blood products | | | | | |
| a) recipient of blood | 0 | 3 (100) | 0 | 0 | 3 (100) |
| b) recipient of clotting factor | 0 | 3 (100) | 1 (1.9) | 0 | 4 (100) |
| Heterosexual contact/endemic | | | | | |
| a) origin in pattern II country | 0 | 1 (8.3) | 8 (66.7) | 3 (25) | 12 (100) |
| b) sexual contact with person at risk | 5 (3.6) | 126 (91.3) | 5 (3.6) | 2 (1.4) | 138 (100) |
| Occupational exposure | 0 | 1 (100) | 0 | 0 | 1 (100) |
| NIR-HET ⁴ | 7 (4.8) | 116 (79.4) | 20 (13.7) | 3 (2.1) | 146 (100) |
| Other | 1 (100) | 0 | 0 | 0 | 1 (100) |
| NIR ⁵ | 4 (1.5) | 255 (95.2) | 7 (2.6) | 2 (0.7) | 268 (100) |
| Perinatal | 1 (100) | 0 | 0 | 0 | 1 (100) |
| Total | 24 (1.8) | 1,222 (93.1) | 54 (4.1) | 12 (0.9) | 1,312 (100) |

¹ Other includes HIV-1 subtypes D, E, A/B, A/C, and A/G. HIV-1 subtype E has also been referred to as the circulating recombinant form (CRF) A/E.

² MSM refers to men who have sex with men.

³ IDU refers to injecting drug use.

⁴ NIR-HET refers non-identified risk related to heterosexual exposure.

⁵ NIR refers to non-identified risk exposures, i.e., when risk exposures were not identified.

Table 19 shows the prevalence of HIV-1 subtypes by ethnicity. While the samples among certain ethnic groups are small and the data are not representative of all newly diagnosed cases of HIV infection, the results suggest that a higher proportion of African/Caribbean people (Black), Asians (including South Asian) and people of mixed ethnicities may be infected with non-B HIV-1 subtypes than the Caucasian (White) population. These results may be due to travel and migration from countries where non-B strains of HIV-1 prevail, but additional data and investigation are required to confirm this hypothesis.

Table 19: Prevalence of HIV-1 subtypes by ethnicity

| Ethnicity | HIV-1 subtype | | | | Total |
|----------------|-----------------|-------------------|-----------------|--------------------|--------------------|
| | A ¹ | B ² | C ³ | Other ⁴ | |
| | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) |
| White | 6 (0.9) | 641 (96.3) | 15 (2.3) | 3 (0.5) | 665 (100) |
| Black | 5 (9.8) | 24 (47.1) | 18 (35.3) | 4 (7.8) | 51 (100) |
| Aboriginal | | | | | |
| Native Indian | 1 (0.8) | 123 (94.6) | 6 (4.6) | 0 | 130 (100) |
| Metis | 0 | 22 (100) | 0 | 0 | 22 (100) |
| Inuit | 0 | 3 (100) | 0 | 0 | 3 (100) |
| Unspecified | 3 (4.7) | 61 (95.3) | 0 | 0 | 64 (100) |
| Asian | 2 (8) | 21 (84) | 1 (4) | 1 (4) | 25 (100) |
| South Asian | 0 | 15 (83.3) | 1 (5.6) | 2 (11.1) | 18 (100) |
| Latin-American | 0 | 19 (100) | 0 | 0 | 19 (100) |
| Other (mixed) | 2 (9.1) | 12 (54.5) | 6 (27.3) | 2 (9.1) | 22 (100) |
| Total | 19 (1.9) | 941 (92.3) | 47 (4.6) | 12 (1.2) | 1,019 (100) |

¹ Ethnicity was unknown for five individuals with HIV-1 subtype A infection.

² Ethnicity was unknown for 281 individuals with HIV-1 subtype B infection.

³ Ethnicity was unknown for seven individuals with HIV-1 subtype C infection.

⁴ Other includes HIV-1 subtypes D, E, A/B, A/C, and A/G HIV-1 subtype E has also been referred to as the circulating recombinant form (CRF) A/E.

Table 20 shows the prevalence of HIV-1 subtypes among recently acquired (within approximately the previous 4 months) versus prevalent infections. The lack of availability of test kits aimed at determining incident infections has affected the extent to which these data could be generated. The sample size is therefore not reflective of all newly diagnosed cases of HIV-1 infection or samples for which HIV-1 subtyping has been completed. Therefore, significant associations between time of infection and HIV-1 subtype could not be determined. However, in samples that have been analyzed for time of HIV infection, the data suggest that HIV-1 subtype C constitutes 3.1% of recent HIV infections. Serologic assays that have been developed to detect recently acquired infections have been based on subtype B derived antigens and have been shown to misdiagnose incident non-B infections as prevalent infections. Further investigation is required to determine the sensitivity of the commercially available assays to accurately detect recently acquired infections in other non-B subtypes of HIV-1.

Table 20: Prevalence of HIV-1 subtypes by recently acquired versus prevalent HIV-1 infections

| HIV-1 infection | HIV-1 subtype | | | | Total |
|-------------------------------|----------------|-----------------|-----------------|--------------------|------------------|
| | A | B ¹ | C | Other ² | |
| | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) | <i>n</i> (%) |
| Recent infection ³ | 0 | 156 (96.9) | 5 (3.1) | 0 | 161 (100) |
| Prevalent infection | 3 (0.7) | 377 (92.9) | 20 (4.9) | 6 (1.5) | 406 (100) |
| Total | 3 (0.5) | 533 (94) | 25 (4.4) | 6 (1.1) | 567 (100) |

¹ Out of 535 HIV-1 subtype B infected individuals who were tested, time of HIV infection could not be determined for two individuals.

² Other includes HIV-1 subtypes D, E, A/C, and A/G.

³ Because of kit availability, a combination of three assays (Organon Technika, Avidity Index, and Abbott) was used to determine recent infections. If any one test identified a sample as being from a recent infection, the sample was included in this category.

Table 21 shows the prevalence of primary drug resistance among HIV-1 subtypes. Since drug resistance genotyping began in 1999, almost 1 year after subtype testing had been initiated, not all samples that have been subtyped have been tested for drug resistance. This implies that samples received before the initiation of drug resistance testing (e.g. all samples from Newfoundland and Labrador) have not yet been tested for drug resistance. Neither are the data representative of all newly diagnosed cases of HIV-1 infection. However, the results indicate that while multi-drug resistant HIV-1 has not been identified in non-B subtypes of HIV-1, primary drug resistance has been identified in subtypes C and A.

Table 21: Prevalence of HIV-1 subtypes by primary drug resistance

| | HIV-1 subtype | | | | |
|----------------------------------------|----------------|------------------|-----------------|--------------------|-------------------|
| | A ¹ | B ² | C ³ | Other ⁴ | Total |
| Drug resistance mutations | <i>n (%)</i> | <i>n (%)</i> | <i>n (%)</i> | <i>n (%)</i> | <i>n (%)</i> |
| Wild type/minor mutations ⁵ | 6 (85.7) | 727 (92.7) | 29 (96.7) | 8 (100) | 770 (92.9) |
| NRTI ⁶ | 0 | 34 (4.3) | 1 (3.3) | 0 | 35 (4.2) |
| NNRTI ⁷ | 0 | 4 (0.5) | 0 | 0 | 4 (0.5) |
| Protease | 1 (14.3) | 13 (1.7) | 0 | 0 | 14 (1.7) |
| MDR ⁸ | 0 | 6 (0.8) | | | 6 (0.7) |
| Total | 7 (100) | 784 (100) | 30 (100) | 8 (100) | 829 (100) |

¹ Out of 11 HIV-1 subtype A infected individuals who were eligible for drug resistance testing, amplification was unsuccessful for one individual.

² Out of 841 HIV-1 subtype B infected individuals who were eligible for drug resistance testing, amplification was unsuccessful for 56 individuals.

³ Out of 35 HIV-1 subtype C infected individuals who were eligible for drug resistance testing, amplification was unsuccessful for five individuals.

⁴ Other includes HIV-1 subtypes D, E, A/C, and A/G. Eight individuals infected with these HIV-1 subtypes were eligible for drug resistance testing.

⁵ Wild type indicates that no major mutations associated with drug resistance were identified. Minor mutations refers to genetic variables not associated with drug resistance.

⁶ NRTI refers to nucleoside reverse transcriptase inhibitor.

⁷ NNRTI refers to non-nucleoside reverse transcriptase inhibitor.

⁸ MDR refers to multi-drug resistance and includes mutations in HIV-1 that are associated with resistance to any two of the three classes of antiretroviral drugs (NRTIs, NNRTIs, and protease inhibitors).

Tables 22 shows the results of univariate analyses to determine factors that are significantly associated with HIV-1 non-B subtype infection. Because of small samples in certain categories and associated low statistical power, any differentiation between no true association and an association that was detected by this analysis could not be made. However, the following observations can be cautiously made. Significantly higher proportions of non-B infections were newly diagnosed during 1996 as compared with 1997 (22.7% versus 7.6% respectively, odds ratio [OR] = 3.6). However, when compared with 1997 the proportion of newly diagnosed non-B infections was significantly lower during 2001 (1.7% versus 7.6% respectively, OR = 0.21). Significantly higher proportions of non-B infections were also observed among females than males (14.7% versus 5.8% respectively, OR = 2.5); among individuals who reported heterosexual contact as their primary exposure factor as compared with male to male sex (17.9% versus 3.0%, respectively, OR = 7.1); and among individuals of Black, Asian or mixed ethnicities as compared with Whites (52.9%, 16.3%, 45.5% versus 3.6% respectively, OR = 30, 5.2, 22.5 respectively).

Table 22.1: Epidemiologic characteristics of individuals infected with non-B subtypes of HIV-1

| | Sample size | HIV-1 non-B | Univariate analysis | |
|--------------------------------------------|-------------|--------------|---------------------|--------------------------------|
| | | <i>n</i> (%) | <i>p</i> value | Crude OR (95% CI) ¹ |
| Year of first diagnosis² | | | | |
| ≤ 1995 | 67 | 4 (6) | 0.77 | – |
| 1996 | 66 | 15 (22.7) | 0.01 | 3.6 (1.4-9.0) |
| 1997 | 105 | 8 (7.6) | Ref | Ref |
| 1998 | 152 | 11 (7.2) | 1 | – |
| 1999 | 342 | 31 (9.1) | 0.84 | – |
| 2000 | 307 | 15 (4.9) | 0.33 | – |
| 2001 | 174 | 3 (1.7) | 0.02 | 0.21 (0.06-0.82) |
| 2002 | 4 | 0 | not done | not done |
| Age (years)³ | | | | |
| < 15 | 6 | 3 (50) | not done | not done |
| 15-19 | 20 | 0 | not done | not done |
| 20-29 | 259 | 26 (10) | 0.15 | – |
| 30-39 | 444 | 31 (7) | Ref | Ref |
| 40-49 | 285 | 20 (7) | 1 | – |
| 50-59 | 86 | 4 (4.7) | 0.63 | – |
| ≥ 60 | 38 | 3 (7.9) | 0.74 | – |
| Gender | | | | |
| Male | 858 | 50 (5.8) | Ref | – |
| Female | 257 | 38 (14.7) | < 0.001 | 2.53 (1.6-4.0) |

¹ Odds ratios were based on a comparison of the variable of interest with the reference (Ref) variable in the group. Only significant ORs are indicated.

² The data from 1996 are largely from B.C., and additional data are needed to interpret the findings from this year.

³ Age reflects age at first diagnosis and is calculated by subtracting year of birth from year at first diagnosis of HIV.

Table 22.2: Epidemiologic characteristics of individuals infected with non-B subtypes of HIV-1 (cont'd)

| | Sample size | HIV-1 non-B | Univariate analysis | |
|--------------------------------------------|-------------|--------------|---------------------|--------------------------------|
| | | <i>n</i> (%) | <i>p</i> value | Crude OR (95% CI) ¹ |
| Exposure category² | | | | |
| MSM | 336 | 10 (3) | Ref | Ref |
| MSM/IDU | 22 | 2 (4.5) | 0.63 | – |
| IDU | 358 | 9 (2.5) | 0.82 | – |
| Blood/blood products | 7 | 1 (14.3) | not done | not done |
| Heterosexual | 296 | 53 (17.9) | < 0.001 | 7.1 (3.5-14.3) |
| Occupational exposure | 1 | 0 | not done | not done |
| Perinatal | 1 | 1 (100) | not done | not done |
| Other | 1 | 1 (100) | not done | not done |
| No identified risk | 268 | 13 (4.9) | 0.29 | – |
| Ethnicity³ | | | | |
| White | 665 | 24 (3.6) | Ref | Ref |
| Black | 51 | 27 (52.9) | < 0.001 | 30 (15.1-59.6) |
| Aboriginal | 219 | 10 (4.6) | 0.54 | – |
| Asian | 43 | 7 (16.3) | 0.002 | 5.2 (2.1-12.9) |
| Latin-American | 19 | 0 | not done | not done |
| Other (mixed) | 22 | 10 (45.5) | < 0.001 | 22.5 (8.8-56.8) |
| Primary drug resistance⁴ | | | | |
| Wild type/minor mutations | 770 | 43 (5.6) | Ref | Ref |
| Major mutations | 59 | 2 (3.4) | 0.74 | – |
| Time of HIV-1 infection⁵ | | | | |
| Prevalent | 406 | 29 (7.1) | Ref | Ref |
| Recent | 161 | 5 (3.1) | 0.08 | 0.41 (0.16-1.1) |

¹ Odds ratios were based on a comparison of the variable of interest with the reference (Ref) variable in the group. Only significant ORs are indicated.

² MSM refers to men who have sex with men, IDU refers to injecting drug use. The heterosexual category consists of heterosexual contact in endemic areas, high-risk heterosexual contact, and non-identified risk related to heterosexual contact.

³ People of Native Indian, Metis, and Inuit origin were grouped under the Aboriginal category. People of Asian and South Asian origin were grouped under the Asian category.

⁴ Wild type indicates that no major mutations associated with drug resistance were identified. Minor mutations refers to genetic variables not associated with drug resistance. Major mutations refers to genetic changes associated with resistance to protease inhibitors and reverse transcriptase inhibitors.

⁵ Because of kit availability, a combination of three assays (Organon Technika, Avidity Index, and Abbott) was used to determine recent infections. If any one test identified a sample as being from a recent infection, the sample was included in this category.

5.0 National HIV Reference Services

5.1 Background

The provincial public health laboratories, the Canadian Blood Services, and HÉMA-QUÉBEC test thousands of samples each year and thus serve as crucial partners in the CHSDRSP. A number of factors can cause serologic tests to yield fallible results: samples originating from seroconverters; samples that are cross-reactive, for example to HIV-2; and, as seen in 1993 with the failure of some diagnostic kits in France to detect HIV-1 subtype O, the appearance of divergent strains of HIV-1. Furthermore, genetic variants of HIV can be problematic for HIV PCR and viral load tests, often leading to discordant findings with serologic testing.

As a part of their reference services, the national HIV laboratories test samples showing unusual serologic, PCR or other virologic test results. This function of the national HIV laboratories is crucial in addressing one of the goals of the CHSDRSP – protection of the blood supply – since screening tests should be able to detect all circulating strains of HIV in Canada. This relationship between the provincial and national laboratories may also serve other programs of the national HIV laboratories, including quality assurance and diagnostic kit monitoring.

As of June 30, 2002, 38 diagnostic samples from untreated individuals from five provinces have been submitted for subtype analysis through the sentinel arm of the CHSDRSP. Results from 25 samples that were successfully amplified and subtyped are shown in Table 23. It is of note that mutation L90M associated with resistance to the protease inhibitors nelfinavir and saquinavir was identified in one sample from Ontario taken from an individual infected with HIV-1 subtype C.

Table 23: HIV-1 subtype distribution of samples submitted to reference services

| Province | HIV subtype (no. samples) | Year of HIV diagnosis |
|---------------------------|-----------------------------------|-----------------------|
| Newfoundland and Labrador | HIV-1 subtype A (1) | 1999 |
| Nova Scotia | HIV-1 subtype C (1) | 1998 |
| Ontario ¹ | HIV-1 subtype B (1) | 1997 |
| | HIV-1 subtype B (1) | 1999 |
| | HIV-1 subtype B (6) | 2000 |
| | HIV-1 recombinant subtype A/G (2) | 2000 |
| | HIV-1 subtype B (5) | unknown |
| | HIV-1 subtype C (3) | unknown |
| | HIV-1 subtype A (1) | unknown |
| | Manitoba | HIV-1 subtype C (1) |
| HIV-1 subtype C (1) | | 1999 |
| HIV-1 subtype B (1) | | 1999 |
| Alberta | HIV-1 subtype B (1) | 1998 |

¹Samples from eight treatment experienced individuals whose infection was diagnosed in Ontario were not included in this table.

5.2 HIV-2 infections

There is currently no active surveillance for HIV-2 in Canada. An informal survey of provincial public health laboratories indicated that between 1988 and November 2002, 56 new cases of HIV-2 infection had been identified in Canada. The majority of these cases were identified in Ontario and Quebec. This number is likely to be an underestimate, since it is very possible that, because the only approved HIV western blot kit is HIV-1 specific, HIV-2 cases have been reported as HIV-1. Discussions are currently under way to explore the need (if any) to enhance surveillance for HIV-2.

Technical Notes

Data collection and reporting

The results presented here represent individuals who sought testing, whose condition was properly diagnosed, and whose results were reported to provincial health authorities and Health Canada. Furthermore, they include only those individuals for whom sufficient sera, taken for the purposes of diagnostic testing, were available to send to the national HIV laboratories and, of these, the subset for whom subtype analysis and/or primary drug resistance genotyping was completed as of June 30, 2002. The quality of samples that are received by the national HIV laboratories also determines whether subtype and primary drug resistance results can be generated. Typically, the laboratories make at least two attempts on samples that are difficult to amplify with the in-house and consensus group M primers. The National Laboratory for HIV Genetics is currently in the process of examining the use of other primer sets for RT-PCR amplification.

The epidemiologic data collected through the CHSDRSP consist of information routinely collected on the national or provincial HIV case reporting forms plus additional data that allow interpretation of the laboratory results. These additional data include type of laboratory specimen sent, date of last negative HIV test, history of seroconversion (if any), antiretroviral treatment history (if any) and viral load count at diagnosis. These enhanced data are usually collected when the affected individual seeks treatment. Not all individuals with a diagnosis of HIV seek treatment; furthermore, linkages with clinical databases may not be feasible to collect these data.

The quality and completeness of epidemiologic data remain problematic (see Data Limitations section), and one of the key roles of federal field surveillance officers is to work with the provincial and territorial health partners to facilitate the collection and timely reporting of these data to Health Canada.

Exposure category hierarchy

HIV cases were assigned to a single exposure category according to an agreed-upon hierarchy of risk factors. The hierarchy is described in more detail in the *HIV and AIDS in Canada Surveillance Report*, available by contacting the Division of HIV/AIDS Epidemiology and Surveillance or electronically at www.hc-sc.gc.ca/pphb-dgsp/hast-vsmt/public_e.html

Analysis of drug resistance

While both genotypic and phenotypic testing methods are well established, each has its limitations. Both tests provide information only on the virus that predominates at the time of sampling and are unable to identify virus that may be present as a result of previous drug exposures or that is present in “quasi” or minority species. This latter point is particularly important, as minority species of virus may become predominant under selective drug pressures that do not completely inhibit viral replication. Both assays are technically difficult to perform when the concentration of virus is < 1,000 copies/mL and may require highly specialized laboratory facilities and personnel. The ability of both assays to quantify resistance to certain drugs has not yet been determined. Phenotypic testing is expensive, at a cost of about US\$800/test. With genotypic testing, repeat analyses may be required since mutations strongly associated with drug resistance continue to be “discovered”, and their complex interactions are only now beginning to be understood. For this reason, the list of mutations associated with drug resistance is updated annually. The analyses that are presented in this report are based on the list of mutations shown in Appendix 2.

Interpretation of drug resistance

The interpretation of genotypic and phenotypic test results for patient care is still uncertain and under active research. The complexity of this task is compounded by several factors: genotypic and phenotypic test results may not correlate with one another; clinical relevance varies from drug to drug; the concentrations at which a drug is ineffective has not been determined *in vivo*; and the extent to which pharmaceutical interactions influence resistance is not well known. It is anticipated that the list of mutations on which the present results are based will change as new information on drug resistance mutations becomes available over time. International expert review panels meet periodically to review the latest laboratory and clinical findings in the development of guidelines for interpreting genotypic and phenotypic drug resistance mutations for clinical management. A similar panel of national experts to advise the CHSDRSP on drug resistant mutations and their interpretation is being assembled.

Data Limitations

The data presented in this report must be interpreted with caution for the following reasons:

- ◆ The data represent cases of newly diagnosed infections in individuals for whom serum specimens and corresponding epidemiologic information are provided to Health Canada from provincial partners participating in the CHSDRSP. The data are based on convenience sampling and therefore do not include all newly diagnosed cases in a given population for any given year. While we do not anticipate any biases introduced as a result of the convenience sampling, we need to bear in mind that the data are not representative of all newly diagnosed cases in the population.
- ◆ The data presented are only from those individuals who are infected with HIV and who then seek testing. They do not represent individuals who do not know their HIV status or choose not to seek testing. Furthermore, they do not represent individuals from whom there is insufficient sample for strain and drug resistance genotyping.
- ◆ Since some mutations conferring resistance may not be stable over time, the ability to detect resistance is highly dependent on the time since drug pressure was withdrawn. In the case of treatment naïve individuals with newly diagnosed infection, time of drug withdrawal is presumably reflected by time since infection. It is therefore possible that mutations associated with drug resistance are no longer detectable in the older, prevalent infections.
- ◆ Wherever possible and before submission of sample and epidemiologic data, provinces participating in the CHSDRSP review and assess the presence of duplicate positive test reports in order that the data accurately reflect the number of new individuals who test positive for HIV infection. However, because of the nature of HIV reporting in certain jurisdictions (e.g. non-nominal reporting), the removal of all duplicates may not have been possible.
- ◆ The data do not include information from two provinces that bear the burden of HIV infections – Ontario and Quebec. Work is already under way on mechanisms to include data from these provinces, and it is anticipated that one or both of them will be included in the next report on strain and primary drug resistance surveillance in Canada.
- ◆ Since this report deals solely with primary drug resistance, analysis was conducted on the laboratory specimens collected from treatment naïve individuals at the time of initial testing for HIV. However, treatment history cannot always be verified. For example, at least 5% of laboratory specimens from British Columbia are likely to have been collected from individuals who have had treatment.
- ◆ The report presents major mutations associated with drug resistance and not minor mutations, which, in combination, may be associated with drug resistance. The prevalence of primary drug resistance is therefore likely an underestimate of the true prevalence.
- ◆ Subtype and drug resistance analyses are conducted on 1053 base pairs in the *pol* gene and reflect what is observed within this small region of the viral genome.
- ◆ At the time of writing, all data had been received retrospectively, so the report actually describes what has happened historically. While these data are still useful in informing program planning and policy formulation, their utility could be enhanced with “real-time” data.

- ◆ Missing or unknown epidemiologic data remain problematic, particularly with respect to information on previous HIV testing, date of first positive HIV test, ethnicity, risk behaviour, CD4 and viral load at diagnosis, and previous antiretroviral treatment. This may have resulted in misclassification (in the variables presented in this report) or inability to assess the association of a variable of interest (e.g. viral load at diagnosis).
- ◆ Wherever possible, epidemiologic data submitted through this enhanced HIV surveillance initiative have been linked to routinely submitted data on HIV infections. However, because of the nature of reporting in certain jurisdictions, this has not always been possible. Therefore, there may be some discrepancies between the epidemiologic data reported in the routine HIV surveillance report when compared with the data in this report.

Appendix 1*

Glossary of terms

Cross-resistance: resistance selected by one drug, which, in turn, confers resistance to one or more drugs not included in the current treatment.

DNA: deoxyribonucleic acid, the genetic material of a cell.

Drug resistance: decreased susceptibility to a drug.

Drug resistance mutation: a change in amino acid associated with increased resistance of HIV to an antiretroviral drug.

Gene: a segment of DNA coding for a particular protein or protein sub-unit.

Genotype: specific sequence of nucleotides that determine the genes of HIV-1.

Genotypic resistance: presence of mutations to nucleotides that increase resistance of HIV to one or more antiretroviral drugs.

Genotypic tests: conducted to determine the presence of mutations in the nucleotide sequence of the viral genome.

HIV: human immunodeficiency virus.

Incidence: the number of new occurrences of a disease in a given population during a specified period of time.

Major mutation: mutation in the viral nucleotide sequence, which, in and of itself, is strongly associated with conferring increased resistance of HIV to an antiretroviral drug.

Minor mutation: mutation in the viral nucleotide sequence, which, in combination with other mutations, confers increased resistance of HIV to a drug.

Multi-drug resistance: increased resistance of HIV to more than one class of drugs.

Mutation: genetic change in the viral nucleotide sequence.

Nucleotide: a monomeric unit consisting of a sugar, phosphate, and nitrogenous base.

PCR: polymerase chain reaction, a molecular technique used to amplify nucleotide sequences.

Phenotype: characteristics and growth properties of HIV-1.

Phenotypic resistance: when four or more times the amount of drug is required to inhibit viral growth by 50% (IC 50).

Phenotypic tests: used to determine the susceptibility of a virus to drug in a virus culture assay.

Prevalence: the number people with the disease in a given population who are alive during a specified period of time.

* Some definitions are adapted from the *HIV and AIDS in Canada Surveillance Report to December 31, 2000*,

<http://www.hc-sc.gc.ca/pphb-dgspsp/publicat/aids-sida/haic-vsac00/index.html> and from the *International Consultation on Monitoring the Emergence of Antiretroviral Resistance* sponsored by WHO, UNAIDS and ISS (October 2000)

http://www.who.int/emc-documents/antimicrobial_resistance/whocdscrdrs200111c.html

Primary resistance: resistance of HIV to antiretroviral drugs seen in individuals who have never before received treatment and so, presumably, have been infected with drug-resistant virus.

Protease: an enzyme that breaks down proteins to their subunits or component peptides.

Recombinant: HIV-1 containing a sequence corresponding to a mixture of more than one subtype in the envelope gene.

Reverse transcriptase: an enzyme that is unique to all retroviruses. It reads the genetic information of the retrovirus, which is RNA, and makes a DNA copy.

RNA: ribonucleic acid, a polymer of nucleotides involved in protein synthesis.

RT-PCR: PCR using the enzyme reverse transcriptase (RT), a molecular technique used to amplify RNA sequence into DNA.

Secondary resistance: increased resistance of HIV to drugs, seen among individuals already receiving treatment (presumably due to treatment failure).

Subtype: also referred to as clade, a group of related HIV variants, classified according to degree of genetic similarity.

Wild type virus: the most commonly occurring form of HIV-1.

Appendix 2

List of major mutations used for this report¹ and corresponding antiretroviral drugs

Major mutations associated with nucleoside reverse transcriptase inhibitors

| Mutation ² | Antiretroviral drugs ³ |
|-----------------------|-----------------------------------|
| M41L | AZT, d4T |
| K65R | ddC, ddl, Tenofovir, Abacavir |
| <i>D67N</i> | <i>AZT, d4T</i> |
| T69D | ddC |
| K70R | AZT, d4T |
| L74V | ddl, ddC, Abacavir |
| <i>Y115F</i> | <i>Abacavir</i> |
| Q151M | AZT, ddC, ddl, d4T Abacavir |
| M184I | 3TC |
| M184V | 3TC, ddC |
| <i>L210W</i> | <i>AZT, d4T</i> |
| T215F | AZT, d4T |
| T215Y | AZT, d4T |
| <i>K219E</i> | <i>AZT, d4T</i> |
| <i>K219Q</i> | <i>AZT, d4T</i> |

² Major mutations that have been added to the consensus list since the last surveillance report of samples received as of June 30, 2001, are indicated in ***bold italics***.

³ Other common names for the nucleoside reverse transcriptase inhibitors include AZT (zidovudine, retrovir); d4T (stavudine, zerit); ddC (zalcitabine, hivid); 3TC (lamivudine); ddl (didanosine videx); abacavir (ziagen); tenofovir (viread).

¹ **Note:** The correlation of drug resistance to genotype in this report is based on scientific consensus of major mutations associated with HIV resistance to antiretroviral drugs as of June 2002. These correlations do not necessarily imply phenotypic resistance to a particular antiretroviral drug in a clinical setting.

**Major mutations associated with
non-nucleoside reverse transcriptase inhibitors**

| Mutation¹ | Antiretroviral drugs² |
|-----------------------------|-----------------------------------------|
| <i>L100I</i> | <i>DLV, NVP, EFV</i> |
| K103N | DLV, NVP, EFV |
| V106A | NVP |
| <i>V108I</i> | <i>NVP, EFV</i> |
| Y181C | NVP, EFV, DLV |
| Y181I | NVP, EFV |
| <i>Y188C</i> | <i>NVP</i> |
| <i>Y188H</i> | <i>NVP</i> |
| <i>Y188L</i> | <i>DLV, NVP, EFV</i> |
| G190A | NVP, EFV |
| G190S | EFV |
| <i>P225H</i> | <i>EFV</i> |
| <i>M230I</i> | <i>NVP, EFV, DLV</i> |
| <i>P236L</i> | <i>DLV</i> |

¹ Major mutations that have been added to the consensus list since the last surveillance report of samples received as of June 30, 2001, are indicated in ***bold italics***.

² Other common names for the nucleoside reverse transcriptase inhibitors include efavirenz (sustiva); DLV (delavirdine, rescriptor); NVP (nevirapine, viramune).

**Major mutations associated with
resistance to protease inhibitors**

| Mutation¹ | Antiretroviral drugs² |
|-----------------------------|-----------------------------------------|
| D30N | nelfinavir |
| M46I | indinavir |
| M46L | indinavir |
| G48V | saquinavir |
| 150V | amprenavir |
| <i>150L</i> | <i>atazanavir</i> |
| V82A | indinavir, ritonavir |
| V82F | indinavir, ritonavir |
| V82S | ritonavir |
| V82T | indinavir, ritonavir |
| 184V | amprenavir, indinavir, ritonavir |
| N88D | nelfinavir |
| L90M | nelfinavir, saquinavir |

¹ Major mutations that have been added to the consensus list since the last surveillance report of samples received as of June 30, 2001, are indicated in ***bold italics***.

² Other common names for the protease inhibitors include amprenavir (agenarase); indinavir (crixivan); ritonavir (norvir); saquinavir (invirase, fortovase).