Clozapine-associated neutropenia in Latin America: incidence report of 5380 Chilean users
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Systematic information about Latino clozapine users is still scarce. Our aim was to evaluate the risk of clozapine-associated neutropenia in a Chilean cohort using the last Food and Drug Administration’s recommendations for clozapine monitoring. Findings should improve clinical practice and promote changes in clozapine guidelines in Latin America. We conducted a retrospective observational study of 5380 Chilean clozapine users that started clozapine treatment between 2003 and 2015. The absolute risk of severe neutropenia was 0.61% (33/5380) with an incidence of 0.086 cases per 100 person-years of follow-up. 87.9% of cases with severe neutropenia appeared during first 18 weeks. Cases of mild neutropenia were 3.9% of total sample and occurred almost constantly without a specific risk time. 77.5% of cases of moderate or severe neutropenia didn’t present an event of mild neutropenia before. 22.8% of clozapine users (1227/5380) discontinued treatment for any cause and 4.2% (225/5380) due to neutropenia in any severity level.

Clozapine-associated neutropenia risk in Latino users is similar than in the rest of the world. The evidence of a very low risk for severe neutropenia and the behaviour of mild neutropenia cases confirm the feasibility of changes in Latin American clozapine guidelines using current Food and Drug Administration’s recommendations as a model.

Introduction
The use of clozapine has historically been limited by the risk of neutropenia and agranulocytosis (Anderman and Griffith, 1977). In 1988, Kane et al., showed clozapine’s effectiveness in the management of treatment-resistant cases (Kane et al., 1988), which led to clozapine being widely used again but under strict pharmacovigilance conditions. These included monitoring with periodic blood tests and discontinuation of the drug in cases of neutropenia/agranulocytosis. The preventive measures worked successfully reducing the expected incidence of agranulocytosis and drug-associated fatalities (Honigfeld et al., 1998). Updated data from a recent meta-analysis by Myles et al. reported an incidence of severe neutropenia (agranulocytosis) of 0.9% and a risk of death associated with neutropenia secondary to clozapine of 0.013% (Myles et al., 2018). Despite these potential side effects, clozapine is currently the first-line option for treatment-resistant schizophrenia patients (Kane et al., 1988; Siskind et al., 2016) and in suicidality associated with schizophrenia and schizoaffective disorder (Meltzer et al., 2003).

Recently, the US Food and Drug Administration (FDA) changed blood monitoring parameters for clozapine use. The new guidance was a response to the underutilization of this medication despite its proven effectiveness, trying to decrease the barriers preventing people to start or remain on clozapine (Sultan et al., 2017). The main changes in the new guidelines were (Clozapine REMS Program, 2015; Bastiampillai et al., 2016): (1) Only the absolute neutrophil count (ANC) is monitored rather than total white blood cell count; (2) patients can continue clozapine if they present a case of mild neutropenia (Table 1), unlike the previous guideline which advised stopping clozapine treatment; (3) clozapine should be discontinued in cases of moderate or severe neutropenia (Table 1); (4) restarting clozapine is allowed even after cases of severe neutropenia, if the benefits of reexposure reasonably outweighed the risks; (5) specific parameters for clozapine users with benign idiopathic neutropenia were established. The advice to continue clozapine use during a mild neutropenia event and the possibility to restart this medication even after a severe neutropenia are probably the two main changes that could stimulate a broader use of clozapine in the US and worldwide.

In Latin America, systematic information about clozapine users is still scarce (Table 2). In a recent report of trends in clozapine use among 17 countries, Bachmann et al. included data from a cohort of Colombian patients (Bachmann et al., 2017). Information from two health
insurance funds of the National Health Insurance System of Colombia (outpatient prescription dispensing) was analysed showing a prevalence of clozapine use of 65.8 per 100 000 persons (age-standardised value). Other important findings were a significant decrease in the clozapine use for the observed period (2006–2014) and a marked peak of clozapine in elderly people (80+ years) with a prevalence of 1200/100 000 for male and 761.9/100 000 for women. A report of Balda et al. in Argentina (Balda et al., 2015) showed the incidence of neutropenia events, without a detailed characterization of clozapine users that did not develop hematological side effects. Across 6 years of observation (2007–2012), this study reported a mean annualized incidence of 0.051 per 100 person-years at risk for severe neutropenia. This cohort was the only Latin American clozapine population included in the meta-analysis of Myles et al. (2018). Up to date (November 2018), no other epidemiological reports are available in the PUBMED database about clozapine use in Latin America. This lack of information could be originated in the differences currently existing between the health systems of the different countries, which in turn impacts on the information and registration systems of the users of clozapine in Latin America. Gather more information of clozapine users should improve the prescription of this antipsychotic in our region. These improvements could include a more widespread use of clozapine and the possibility of changes in the regulations of hematological monitoring.

Clozapine has been used in Chile since 1991. In 2000, it was published a Technical Guideline for the Use of Clozapine that established pharmacovigilance requirements for Chilean patients (Ministerio de Salud Chile, 2000). Like other international guidelines, it established that clozapine users should be monitored weekly with blood tests during the first 18 weeks of treatment and then monthly while they were receiving the medication. Clozapine was advised to be discontinued when the neutrophil count fell below 1500/mm³ (mild neutropenia). Echoing FDA’s changes, a new Chilean Clozapine Guideline was launched in October 2018. This new version preserved the frequency of blood monitoring and adopted the neutropenia definitions and FDA’s recommendations for discontinuation of clozapine (Ministerio de Salud. División de Salud de Personas, 2018). As in other countries, a centralized National Clozapine Pharmacovigilance System has followed up clozapine users from the Chilean public health system. An adequate analysis of the records of this Pharmacovigilance System could help to reduce the gap about the information available of clozapine users in Latin America.

Therefore, the aim of the present study is to evaluate the risk of neutropenia secondary to clozapine in a relatively understudied Latino population. The results will raise evidence about the behaviour of Latino users of
clozapine and also around the current FDA’s recommendations and its benefits for clozapine monitoring.

Material and methods

Study population

We included 5380 clozapine users over 16 years old of the Chilean public health system registered in the National Clozapine Pharmacovigilance System between 1 January 2003 and 31 December 2015.

Most cases included had a treatment-resistant schizophrenia diagnosis. The national electronic registry of the Clozapine Pharmacovigilance System has sociodemographic data of the patients and the records of the blood tests performed in the analyzed period. More than 90 mental health teams in the public health system of Chile prescribe clozapine and perform the registration and monitoring of their patients.

Study variables

The sociodemographic variables evaluated were age at treatment initiation, sex, dose of clozapine used, number of weeks in treatment until the suspension or end of the observation period. The follow-up of blood tests was carried out until 31 December 2016, which ensured that all the recruited patients had a minimum of 1 year of follow-up. Discontinuation date was established as the last blood test entered in the electronic record of patient monitoring.

The operational definitions for the cases of mild, moderate and severe neutropenia (agranulocytosis) are summarized in Table 1. Criteria established in the literature and in the new guidelines for clozapine use in the US were used (Gibson and Berliner, 2014; Palmblad et al., 2014; Clozapine REMS Program, 2015). Cases of neutropenia were identified by blood tests results. For each case of neutropenia, we analyzed: age at treatment initiation, age at the time of the episode of neutropenia, sex, dose of clozapine used, number of weeks in treatment and time of clozapine suspension, if it happened.

Statistical analysis

Categorical variables were reported as frequency and percentage and were compared using contingency tables (Chi-square). Continuous variables were evaluated according to graphical methods and with the Kolmogórov–Smirnov or Shapiro–Wilk test, as appropriate; they were reported as means ± SD and medians and were compared using ANOVA (t-test) or the Mann–Whitney U test for non-normal distributions of continuous variables. Logistic regression to analyse covariables was used. Time of onset for hematological alterations was estimated using Kaplan–Meier survival analysis and compared with the Log-Rank test. The level of significance was established with a value \( P < 0.05 \). SPSS 15 (version 15.0; SPSS Inc., Chicago, IL) was used for the statistical analysis.

This study was approved by the ethics committee of the North Metropolitan Health Service, Santiago, Chile.

Results

Clozapine users and discontinuation of treatment

The main characteristics of the 5380 clozapine users registered between 2003 and 2016 are shown in Table 3. There were more significantly more men (64.1%) than women (35.9%) on treatment with clozapine (\( P < 0.0001 \), Chi-square). Men were significantly younger at clozapine initiation than women [mean 31.6 years old (95% confidence interval [CI]: 31.3–32) and 36.4 years (95% CI: 35.8–37), respectively; \( P < 0.0001 \), Mann–Whitney]. Likewise, men used higher doses of clozapine in comparison with women [mean 344.9 mg/day (95% CI: 339–350.8) and 298.5 mg/day (95% CI: 291.2–305.9), respectively, \( P < 0.0001 \), Mann–Whitney].

The median of follow-up time for all the sample was 382 weeks [interquartile range (IQR): 158–589]. 22.8% of clozapine users (1227/5380) discontinued treatment for any cause, with significantly higher rates among women in comparison with men. 4.2% of the total sample (225/5380) discontinued their treatment due to neutropenia (any severity) with significantly higher rates among women in comparison with men (Table 3). With a total of 38269.3 person-years of follow-up, we observed an incidence rate of discontinuation for any cause of 3.21 per 100 person-years and 0.58 per 100 person-years for discontinuation related to neutropenia (at any level of severity).

Table 3 Description of sociodemographic variables and causes of discontinuation of clozapine users at the national level (2003–2016) in Chile, by sex

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>Total</th>
<th>Male</th>
<th>Female</th>
<th>( P )-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Clozapine users 2003–2016, N (%)</td>
<td>5380</td>
<td>3451 (64.1)</td>
<td>1929 (35.9)</td>
<td>&lt;0.0001 ( a )</td>
</tr>
<tr>
<td>Age, years; mean (SD/median IQR)</td>
<td>33.3 (12.3)/32 (23–42)</td>
<td>31.6 (11.5)/29 (22–40)</td>
<td>36.4 (13.2)/36 (26–46)</td>
<td>&lt;0.0001 ( * )</td>
</tr>
<tr>
<td>Doses, mg; mean (SD/median IQR)</td>
<td>328.3 (170.8)/300 (200–400)</td>
<td>344.9 (173.8)/300 (200–450)</td>
<td>298.5 (160.3)/300 (200–400)</td>
<td>&lt;0.0001 ( * )</td>
</tr>
<tr>
<td>Neutropenia associated discontinuation, N (%)</td>
<td>225 (4.2)(^ b )</td>
<td>127 (3.7)</td>
<td>98 (5.1)</td>
<td>0.003(^ * )</td>
</tr>
<tr>
<td>Other causes discontinuation, N (%)</td>
<td>1002 (18.6)(^ c )</td>
<td>616 (18.5)</td>
<td>386 (21.1)</td>
<td>0.027(^ b )</td>
</tr>
</tbody>
</table>

\( \text{IQR, interquartile range.} \)

\(^ {a} \text{Mann–Whitney U.} \)

\(^ {b} \text{Chi-square.} \)

\(^ {c} \text{Percentage in relation to total users analysed (cumulative incidence).} \)
Rates of different levels of neutropenia and mortality

Table 4 shows clozapine users without neutropenia (controls) and with different levels of neutropenia (cases) according to sex, age, and last registered doses.

The cumulative incidence of severe neutropenia (ANC < 500/µl) was 0.61% (33/5380). The incidence rate was 0.086 cases per 100 person-years of follow-up. All those who presented it stopped the treatment. There was no difference in gender distribution and clozapine’s doses between severe neutropenia cases and controls. In terms of age, the logistic regression showed that the group over 45-year-old had more risk to develop severe neutropenia in comparison with patients under 45-year-old [odds ratio (OR): 2.54; 95% CI: 1.23–5.25; \( P = 0.012 \)] adjusted by sex and doses.

The cumulative incidence of moderate neutropenia (ANC 500–999/µl) was 1.28% (69/5380). The incidence rate was 0.18 cases per 100 person-years of follow-up, and all of them stopped clozapine treatment. There was no difference in gender distribution and clozapine doses between moderate neutropenia patients and controls. In terms of age, the logistic regression showed that the group over 45-year-old had more risk to develop moderate neutropenia in comparison with patients under 45-year-old (OR: 2.07; 95% CI: 1.23–3.48; \( P = 0.006 \)) adjusted by sex and doses.

The cumulative incidence of mild neutropenia (ANC 1000–1499/µl) was 3.9% (209/5380). The incidence rate was 0.55 cases per 100 person-years of follow-up, and 123 (58.9%) patients discontinued clozapine. There was no difference in gender distribution between mild neutropenia patients and controls. The logistic regression showed that group over 45 years did not have more risk to developing mild neutropenia in comparison with patients under 45-year-old (OR: 0.67; 95% CI: 0.44–1.012; \( P = 0.057 \)) adjusted by sex and doses.

Four clozapine users died due to infections presented in the context of severe neutropenia (cumulative incidence of 0.074%). The incidence rate of mortality associated with neutropenia is 0.01 per 100 person-years of follow-up. Table 5 summarizes the characteristics of the cases of deceased patients.

Risk of different levels of neutropenia: survival analysis

Fig. 1 shows the Kaplan–Meier curve of the time of appearance and discontinuation of treatment due to neutropenia events according to severity of the neutropenia. The median to present severe neutropenia was 9 weeks (IQR: 6.5–12.5), and 87.9% of cases appeared during the first 18 weeks. The median to present moderate neutropenia was 16 weeks (IQR: 11–42), and 58% of cases appeared during the first 18 weeks. The median to present mild neutropenia was 282 weeks (IQR: 36–514), and 27.6% of cases emerged in the first 18 weeks. When
looking at the time of appearance of severe, moderate and mild neutropenia, there is a statistically significant difference between the analyzed curves (Test Log-Rank: Chi-square = 16.32 for ANC < 500/µl; Chi-square = 70.221 for 1000 > ANC ≥ 500/µl; Chi-square = 107.035 for ANC 1000 ≤ ANC < 1500/µl; P value <0.0001). Only 22.5% (23/102) of clozapine users that presented a severe and moderate neutropenia had a previous episode of mild neutropenia. Therefore, most cases (77.5%) had a severe or moderate neutropenia without a previous warning signal.

**Discussion**

This report presents a Latin American cohort of clozapine users and their risk of neutropenia and discontinuation of treatment. The absolute risk of severe neutropenia (ANC < 500/µl) was 0.61% (33/5380) with an incidence rate of 0.086 cases per 100 person-years of follow-up. Most of the moderate and severe cases of neutropenia emerged during the first 18 weeks of treatment causing the interruption of clozapine treatment. The cases of mild neutropenia were the most frequent (3.9% of total sample) and appeared at any time of treatment. In this group, discontinuation occurred in nearly 60% of the cases. Most of clozapine users (77.5%) with moderate or severe neutropenia did not present an event of mild neutropenia before their more severe lowering in neutrophil count. Globally, 22.8% of clozapine users (1227/5380) discontinued treatment for any cause and 4.2% (225/5380) due to neutropenia in any severity level.

Rates of neutropenia found in our cohort were similar to those reported in previous studies. The cumulative incidence for severe (0.61%), moderate (1.28%) and mild (3.9%) neutropenia closely resembles a recent meta-analysis that reported incidences of 0.9% for severe neutropenia, 1.3% for moderate neutropenia and 3.8% for mild neutropenia (Myles et al., 2018). As such, our patients behave similarly to other groups in the world (Alvir et al., 1993; Munro et al., 1999; Lambertenghi Deliliers, 2000; Lahdelma and Appelberg, 2012), and idiosyncratic factors such as genetic background or type of clozapine prescribed do not seem to have a big effect. The fact that we decided to use only the FDA definitions for the different levels of neutropenia, was an important difference with the previous report of Balda et al. in Latin American population (Balda et al., 2015) and could allow a more easily inclusion of this population in future metanalysis. Severe neutropenia events (agranulocytosis) occurred mostly

### Table 5 Characteristics of fatalities related to severe neutropenia (agranulocytosis)

<table>
<thead>
<tr>
<th></th>
<th>Sex</th>
<th>Age</th>
<th>Week of onset of severe neutropenia</th>
<th>Last dose of clozapine registered (mg/day)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Case 1</td>
<td>Male</td>
<td>21</td>
<td>8</td>
<td>300</td>
</tr>
<tr>
<td>Case 2</td>
<td>Male</td>
<td>16</td>
<td>16</td>
<td>300</td>
</tr>
<tr>
<td>Case 3</td>
<td>Male</td>
<td>74</td>
<td>13</td>
<td>150</td>
</tr>
<tr>
<td>Case 4</td>
<td>Female</td>
<td>65</td>
<td>9</td>
<td>175</td>
</tr>
</tbody>
</table>

**Fig. 1**

Kaplan–Meier curve for the time of appearance of the different levels of neutropenia in clozapine users in Chile between 2003 and 2016.
before 18 weeks, which confirms the need to maintain intensive hematological surveillance during this period. The survival analysis of the three levels of neutropenia showed with Kaplan–Meir curves highlights the differences between the time of appearance and risk of different types of neutropenia across time and reinforce the fact of the different behaviour of the mild clozapine-associated neutropenia events. The increasing risk for severe neutropenia with age has been reported previously (Alvir et al., 1993; Atkin et al., 1996; Munro et al., 1999). Our findings remark a higher risk of severe neutropenia in the group over 45-year-old, a fact that must be considered by clinicians specially when they consider to start clozapine in old age cases (>65 years).

The four cases of death associated with severe neutropenia secondary to clozapine were identified from the registration system. The cumulative incidence rate of 0.074% is higher than 0.013% reported in the Myles meta-analysis already mentioned. Although it is a low incidence of cases, its existence reinforces the need to maintain an active surveillance of neutropenia research, especially during the first treatment period.

The previous report of Balda in Argentina (Balda et al., 2015) showed the sociodemographic characteristics of 38 clozapine users that developed severe neutropenia. We complement that information with the description of a whole population of 5380 clozapine patients. The cohort here presented had a similar sex distribution compared to the reported by Munro in the UK almost 20 years ago by (Munro et al., 1999). The highest proportion of men (64.1%) could explain the higher frequency of discontinuation of treatment for all causes among men (Table 2). Our cohort is younger (average age 36.4 years for women, 31.6 years for men) than the reported by Munro (average age 39.7 years for women, 35.4 years for men). This could be taken as an indirect indicator of a faster use of clozapine in cases of treatment-resistant psychosis among Chilean clinicians. An early use of clozapine in Latino patients with a diagnosis of schizophrenia was already reported for our group (Mena et al., 2018). In that Chilean cohort of 1195 patients with a first episode of schizophrenia, the highest probability of being prescribed clozapine was during the first year of treatment (probability of 0.11, 95% CI: 0.093–0.13), with a median age of clozapine users of 24 years (20–32). Future research in Latino treatment-resistant psychosis population should consider these findings in order to compare clozapine prescription among different countries.

The discontinuation rate is a relevant finding in our cohort. After 1 year of starting clozapine, 75% of the patients will continue on treatment. This information is relevant for prescribers and clozapine users especially because failures in adherence to treatment have been reported as a major problem in the chronic use of antipsychotics reaching up to 74% in a 18 months follow-up period (Lieberman et al., 2005). Our data confirm the advantage of clozapine over other antipsychotics in terms of adherence, a fact previously reported in smaller cohorts of clozapine users (McEvoy et al., 2006).

The results observed raise new evidence around the FDA’s recommendations for clozapine monitoring. Few reports are available since the last update of this guidelines in 2015 implementation (Sultan et al., 2017). Data here presented are especially relevant for one of the main changes of the FDA’s guidance that is keeping patients on clozapine when a mild neutropenia event occurs. Our results showed that mild neutropenia behaves differently from moderate and severe episodes, supporting the idea that it is not necessarily part of a continuum (Bastiaanpillai et al., 2016). Unlike moderate and severe neutropenia, mild neutropenia events did not cluster in the first year of exposure to clozapine (Fig. 1). On the other hand, mild neutropenia could not be established as a clear risk factor for a more severe level of neutropenia, with most of a moderate and severe neutropenia appearing without a mild neutropenia event beforehand (77.5%). From a clinical point of view, these findings should encourage to restart clozapine in patients who have previously discontinued treatment with clozapine for episodes of mild neutropenia or allow the new patients to be maintained on clozapine despite a mild lowering in the neutrophil count.

Our study has several strengths. The number of patients included in the follow-up puts it within the 10 largest cohorts reported in the literature. Patients also had a minimum follow-up time of 1 year. Furthermore, our data comes from ‘real-world’ patient records within a relatively under-funded health system, which increases its translational value to middle- and lower-income countries. Weaknesses that should be considered are related mainly to the lack of data included in other reports such as the incidence of infections in cases of severe neutropenia or the use of other concomitant drugs. Although there is a national regulation regarding the mandatory reporting of clozapine users’ data, there is a risk of under-report, especially in cases of suspension of treatment due to non-hematological causes.

In conclusion, information obtained in this study confirms the safety of clozapine use in Latino population in relation with the risk of neutropenia and fatalities associated. The main findings are encouraging in terms of the feasibility of renewing monitoring guidelines for Latin America in a similar way than the US-FDA. These changes must include, at least, the blood monitoring only with the ANC, continuation of clozapine in case of mild neutropenia and the discontinuation of clozapine in cases of moderate or severe neutropenia. The new guidelines available in Chile since 2018 could be also a good model for other countries of the region in this update process.
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Conflicts of interest
There are no conflicts of interest.

References


