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TITLE PAGE

Title:

Amyotrophic lateral sclerosis as an adverse drug reaction: a disproportionality analysis of the Food and Drug Administration Adverse Event Reporting System

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ABSTRACT

INTRODUCTION

Amyotrophic lateral sclerosis (ALS) is a fatal progressive disease with a still unclear multifactorial etiology. This study focused on the potential relationship between drug exposure and the development of ALS by performing a detailed analysis of events reported in the FDA Adverse Event Reporting System (FAERS) database.

METHODS

The FAERS quarterly data (January 2004-June 2020) were downloaded and deduplicated. The reporting odds ratios (RORs) and their 95% confidence intervals (95%CIs) were calculated as a disproportionality measure. The robustness of the disproportion was assessed accounting for major confounders (i.e., using a broader query, restricting to suspect drugs, and excluding reports with ALS as an indication). Disproportionality signals were prioritized based on their consistency across analyses (ROR stability).

RESULTS

1188 ALS cases were retained. Sixty-two drugs showed significant disproportionality for ALS onset in at least one analysis, and 31had consistent ROR stability, including TNF-α inhibitors and statins. Disproportionality signals from ustekinumab, an immunomodulator against interleukins 12-23 used in autoimmune diseases, and the anti-IgE omalizumab were consistent among analyses and unexpected.

CONCLUSIONS

For each drug emerging as possibly associated with ALS onset, biological plausibility, underlying disease and reverse causality could be argued. Our findings strengthened the plausibility of a precipitating role of drugs primarily through immunomodulation (e.g., TNF-α, ustekinumab, omalizumab), but also by impacting metabolism and the musculoskeletal integrity (e.g., statins and bisphosphonates). Complement and NF-kB dysregulation could

represent interesting topics for planning translational mechanistic studies on ALS as an adverse drug effect.

KEY POINTS

- Amyotrophic lateral sclerosis (ALS) has a complex, unclear etiology including genetic susceptibility and environmental factors.
- By analyzing reports from the FDA Adverse Event Reporting System, 31 drugs were found to be disproportionally reported with ALS, including drugs potentially implicated in ALS development (e.g., immunomodulators, statins).
- Future translational research should focus on the possible role of complement and NF-kB dysregulation in ALS onset or worsening.

MAIN TEXT

1. INTRODUCTION

Amyotrophic lateral sclerosis (ALS) is a rare, subtle, and progressive disease beginning with focal weakness, then spreading to involve most muscles. Patients die due to respiratory paralysis when the diaphragm is involved, in 3 to 5 years from symptoms onset. Disease progression is highly variable among affected individuals, and clinical features encompass heterogeneous motor and nonmotor signs and symptoms [1]. Therefore, late diagnosis is common.

Medications to relieve symptoms are available, together with two specific agents, riluzole and edaravone, which demonstrated a survival benefit of months.

One of the reasons accounting for the failure of almost all previous attempts with clinical trials in ALS is the incomplete comprehension of the pathogenic processes that cause disease onset and progression. Although ALS is a clinically defined syndrome where upper and lower motor neurons degenerate, the pathogenesis is probably heterogeneous across individual cases. It has been suggested that multiple events need to befall, or multiple factors need to be present for the disease to manifest. Presumably, these would include genetic susceptibility and environmental factors. Nonetheless, no specific environmental factor has been proven to cause ALS, and there is a remarkable gap between the relative abundance of proposed genetic ALS determinants and the lack of findings on environmental risk. Of note, the two drugs used to prolong life in patients diagnosed with ALS are thought to act directly on the survival of the motor neurons: edaravone as a free radical scavenger that prevents oxidative stress damage [2], riluzole inhibiting the glutamatergic excitotoxicity that results in high levels of intracellular calcium and free radicals generation [3].

Drugs exposure has been investigated as a potential risk/precipitating factor in the occurrence of ALS. Population-based observational studies (case-control design) found no association with proton pump inhibitors, an association with antibiotics, contradictory results for statins and tumor necrosis factor-alpha (TNF-α) inhibitors, and a potential inverse association with antidiabetics [4-7]. Of note, gut microbiota, altered by several of these drugs, was proposed as a working hypothesis [8].

Large-scale post-marketing databases are suitable sources to suggest potential associations between specific drug exposures and rare events, such as ALS, which may escape detection and/or reporting from randomized controlled trials and population-based studies [9]. Of note, real-world pharmacovigilance databases have been used to investigate the role of drugs in modulating complex and multifactorial neuropsychiatric diseases such as multiple sclerosis, thus supporting clinicians in real-life risk-benefit evaluation [10].

The aim of this study is to characterize spontaneous reports of ALS using one of the largest publicly available repositories of adverse events, namely the FDA Adverse Event Reporting System (FAERS), in order to assess whether specific drug exposures could represent a potential risk or precipitating factor in ALS development.

2. METHODS

2.1 Study design and Data source

The study was conceived as a case/non-case analysis using the FAERS, a surveillance system collecting reports submitted on a voluntary basis by healthcare professionals and consumers. Reports may be submitted directly to the FDA using MedWatch forms but are usually submitted first to the Marketing Authorization Holders, for which forwarding to the FDA is mandatory. It currently collects more than 20 million reports worldwide (including European reports potentially related to serious events and other non-US non-European data), with public

data availability since 2004, thus offering an emerging opportunity for signal detection and characterization [9]. Adverse events and indications are coded using the Medical Dictionary for Regulatory Activities (MedDRA®) preferred terms (PTs). Drug names, instead, are recorded as a free text and are, therefore, extremely heterogenous (e.g., referring to the brand or the active ingredient, not uncommonly with misspellings).

We downloaded the FAERS quarterly data (January 2004-June 2020) and pre-processed them for drug name standardization. We used the WHOdrug dictionary (accessed in March 2020) and manual integration to reach a coverage of more than 97% of the recorded drugs, which were translated into their active ingredients and referred to their class in the Anatomic Therapeutic Chemical (ATC) classification. Further, a semiautomatic algorithm was used to keep only the last update of each report and to remove duplicates (i.e., reports with the same sex, age, weight, country, event date, list of drugs, and list of events) [11].

No ethics board was needed since this was a pharmacovigilance analysis using an existing database with anonymous public data availability.

2.2 Disproportionality analyses

A pre-specified multi-step strategy was applied to assess the consistency of findings and minimize bias.

To evaluate if, and to what extent, ALS was reported as a potential adverse drug reaction, we performed a disproportionality analysis, a consolidated approach in pharmacovigilance. Through the so-called case/non-case approach, the reporting odds ratio (ROR) was calculated and deemed statistically significant when the lower limit of its 95% confidence interval (95%CI) exceeded 1 [11]. If the proportion of ALS reports was greater among patients exposed to a specific drug (cases) than among patients not exposed to it (non-cases), an association was hypothesized (disproportionality signal).

Notably, the performance of the ROR (i.e., the capacity to discriminate true from false positive drug–event associations) is striking especially for adverse events with low/rare background incidence such as ALS [12].

As the main analysis, we calculated the ROR between the PT "amyotrophic lateral sclerosis" and individual active ingredients. In this analysis, we recognized a report as a case both whether the drug was recorded as the suspect therapy or whether it was recorded as concomitant therapy. This choice was made because it may be difficult to identify the culprit for the development of a plausibly delayed onset reaction as ALS, and because, if we are considering concomitant therapy as non-case, then the same drug would appear both at the numerator and the denominator of the ROR formula.

To assess the robustness of the disproportionality signal, we performed three other disproportionalities. To increase the sensitivity of our study we considered 1) a broader ALS definition for case retrieval, including also "familial ALS", "hereditary ALS", "motor neuron disease", "progressive bulbar palsy", and "progressive muscular atrophy". This way we could account for the redundancy in MedDRA terms, for which ALS may be recorded using different terms depending on the reporter. To increase the specificity of the study we restricted the disproportionality analysis to 2) drugs recorded as primary or secondary suspects, and 3) reports without ALS among the specified indications. This way we could account for the judgment of the reporter on the drug role, and for the expected association between ALS and drugs used in its management. In particular, we decided to exclude drugs used to treat ALS only in a specific analysis because the reverse causality bias (i.e., the fact that a disproportion may be the result of drugs being taken to treat ALS or its symptoms) cannot be completely foreseen. This is true, in particular, for drugs used to treat ALS prodromes, which are less specific and develop before the diagnosis.

To further reduce the likelihood of false positives, the ROR was calculated only for drugs with at least 5 cases.

Disproportionality signals were prioritized based on their consistency across the different analyses (ROR stability). Finally, aiming for maximum specificity, we focused on the drugs with the top ROR stability: all the analyses detected a significant ROR.

3. RESULTS

Overall, 14,526,399 reports were downloaded from the FAERS quarterly data and reduced to 10,760,574 with the de-duplication process. Among these, 1,181 reports specified ALS as a reaction (1,464 using the broader definition), 543 (44%) recording ALS as the only adverse event. The geographical areas contributing with more reports were North America (405, 34.1%), Europe (181, 15.2%), Asia (60, 5.1%), and South America (23, 1.9%).

ALS reports occurred more commonly in males, peaked between 50 and 74 years old, and were submitted after 2012.

Sixty-two drugs were disproportionally reported with ALS using the broad definition. Of these, 31 drugs kept the signal in all more specific analyses, 5 drugs in 3 and 17 drugs in 2 (see Table 1). For further information about case numbers and RORs calculated in the analyses, see the supplementary table in the Online Resource.

Some pharmacological classes included more drugs disproportionally reported with ALS. Concerning cardiovascular/metabolic drugs, all marketed statins showed a “top” ROR stability (4 out of 4 analyses) and a high number of cases (atorvastatin, 154 –main ROR = 4.97, 95%CI = 4.19-5.88 –; simvastatin, 95 – 4.01, 3.25-4.94 –; rosuvastatin, 58 – 4.34, 3.33-5.65–). Only a small decrease in cases was observed in the more specific analyses. As for other drugs in the cardiovascular area, atenolol, enalapril, and candesartan were recorded in a non-negligible number of cases (respectively, 27, 15, 19), but with a lower ROR stability.

Concerning immunomodulating agents, TNF-α inhibitors showed the highest number of cases (adalimumab, 99 –2.03, 1.65-2.49–; etanercept, 89 –1.76, 1.42-2.19–; infliximab, 78 –5.46, 4.34-6.87–). Notable findings were also obtained for ustekinumab (top ROR stability, with 10 cases –2.04, 1.10, 3.81–) and omalizumab (top ROR stability with 8 cases –2.06, 1.03-4.12–).

Among antineoplastic agents, methotrexate (2.46, 1.97-3.09) and rituximab (3.3, 2.37-4.59) had the top ROR stability and the highest number of cases. However, the 99 broad cases of methotrexate decrease to 41 when restricting to suspect drugs and cases without ALS in the indications.

Regarding musculoskeletal system drugs, we observed top ROR stability for risedronic acid (7 –2.57, 1.22-5.41–), baclofen (28 –3.61, 2.48-5.26–), and celecoxib (19 –1.91, 1.21-3–). Among nervous system drugs, only duloxetine showed a top ROR stability.

Edaravone and riluzole, the only two drugs labeled for ALS treatment, showed a disproportionality signal only in the first three case definitions, losing it in the correction for the reverse causality bias.

We also observed top ROR stability for dextromethorphan (17 –7.51, 4.65-12.12–).

Three agents belonging to different therapeutic classes, but with a common quinolinic chemical structure and antinicotinic activity, emerged with not negligible cases and disproportionality signal: quinidine (16 –40.18, 24.51-65.88–, top ROR stability), hydroxychloroquine (19 –2.11, 1.34-3.32–, top ROR stability) and quinine (3 –3.21, 1.03-9.97–, Intermediate ROR stability).

4. DISCUSSION

We investigated ALS from a pharmacovigilance perspective and found a higher-than-expected reporting (disproportionality signal) for 62 different drugs, especially TNF-α inhibitors and statins, thus supporting the hypothesis that drug exposure can have a role in ALS onset. For every single drug or drug class, the biological plausibility of the adverse drug effect was discussed, accounting for a potential reverse causality bias between indication for use and ALS.

Role of immunomodulators

Immunomodulator agents had the highest number of signals, especially TNF-α inhibitors (adalimumab, infliximab, etanercept, abatacept). They are usually prescribed to treat chronic inflammatory/autoimmune diseases (e.g., rheumatoid arthritis and psoriasis), although some preliminary studies proposed them even as a potential treatment for ALS, due to their ability in reducing microglial inflammation [13].

As a matter of fact, specific autoimmune mechanisms responsible for the loss of motoneurons have been advocated as factors promoting ALS. In particular, complement dysregulation and deposition have been identified at the neuromuscular junction prior to nerve cell death in ALS patients [14]. The role of complement in ALS was confirmed in animal models [15,16] and indeed two randomized clinical trials on complement inhibitors (ravulizumab and pegcetacoplan) in ALS patients are currently ongoing [17,18]. Since upregulation of TNF-α causes complement induction, TNF-α inhibitors could hamper ALS progression.

On the other hand, TNF-α inhibitors may also represent a risk factor for ALS onset and progression [13]. This “Janus effect” results from their ability to both activate the PI3K/Akt pathway and inhibit the NF-kB-dependent pathways, which are the major survival pathways in motor neurons [19]. Clinical evidence is also conflicting: some reports, indeed, describe ALS onset in patients with autoimmune diseases treated with TNF-α inhibitors (adalimumab) [20], but a population-based cohort study did not find an increased incidence of ALS in patients with rheumatoid arthritis, regardless of treatment with TNF-α inhibitors [7].

Other immunosuppressants also emerged. The signal of ustekinumab, a fully-humanized monoclonal antibody that binds the p40 subunit of unbound Interleukin (IL) 12 and 23, used in inflammatory bowel diseases and psoriasis, found support in a case report where its use anticipated ALS onset [21]. The signal of omalizumab, an IgE antagonist monoclonal antibody mainly used in severe asthma and allergic respiratory diseases, seems completely unexpected. The signal of hydroxychloroquine, frequently used for its immunosuppressive properties (e.g., in rheumatoid arthritis), is partly supported by evidence on other hydroxychloroquine-induced neuromuscular disorders (i.e., myasthenia gravis) [22].

Our findings from the FAERS suggest that a patient who starts receiving immunomodulating drugs to treat autoimmune diseases may have a greater susceptibility to ALS either for the drug or for the underlying autoimmunity. Going forward, translational medicine studies and larger cohort studies should be carried on, in order to further investigate this relationship between each specific agent (or the whole pharmacological class) and ALS development.

Role of antineoplastic agents

Concerning antineoplastic drugs, we observed statistically significant data for imatinib, rituximab, and methotrexate, the most commonly prescribed drugs of their respective class. Other agents with the same mechanisms could share this effect, but their less common use probably limited the number of reports and hampered the achievement of significant disproportionality.

Rituximab's mechanism of action includes direct effects with complement-mediated cytotoxicity together with antibody-dependent cell-mediated cytotoxicity [23]. Furthermore, treatment with methotrexate is associated with systemic complement activation [24]. The complex activities of these drugs leading to complement activation may be implicated in ALS pathogenesis.

Of interest, the case of imatinib, an oral tyrosine kinase inhibitor (TKI) targeting c-KIT, which also inhibits TNF-α production and could reduce the survival of astrocytes, similarly to other TNF-α inhibitors [25]. Masitinib, analog to imatinib with higher selectivity and potential better safety profile, was found to reduce neuroinflammation by targeting mast cells and neutrophils in pre-clinical models [26] and is now tested for ALS treatment in a phase III multicenter international RCT after a promising phase II study [27].

Overall, these data suggest a possible role of the drugs, rather than of the underlying diseases, as risk factors for ALS, and their multifaceted mechanisms of action deserve to be better explored in relation to ALS with preclinical and clinical studies.

Role of agents for metabolic-cardiovascular disorders

Top ROR stability for all statins emerged from our analyses. Statins are widely prescribed drugs and are generally considered safe medications, except for rare side effects such as myositis. As a matter of fact, according to Golomb B. et al. [28], we found higher disproportionality for lipophilic statins, for which the incidence of rhabdomyolysis is 4 times higher than for monotherapy with pravastatin and fluvastatin [29]. It could be argued that rhabdomyolysis (as well as the rare peripheral neuropathy [30]), caused by lipophilic statins, may uncover the onset of ALS.

On the other hand, high plasma cholesterol levels have been suggested to be neuroprotective for ALS and to be associated with an increased survival time. Statins, by lowering cholesterol inhibiting HMG-CoA reductase, may accelerate functional decline or clinical onset in patients inclined to ALS. Patients on statin therapy also reported a significant increase in muscle cramp frequency and severity [31].

By considering all these aspects, we can argue that statins may unmask preexisting ALS or worsen the disease. Given the milder muscular effects of ezetimibe [32], our findings on this drug may be due to a previous or concomitant use of statins.

As for antihypertensives, we found disproportionality for only a few drugs, with a strong decrease in cases in the more sensitive analyses. Published evidence on patients with ALS from clinical registries provides controversial findings on the impact of cardiovascular disorders on ALS course: Moglia C. et al. observed that arterial hypertension, type 2 diabetes, and cardiovascular risk factors do not influence ALS phenotype and prognosis [33]; while Mandrioli J. et al., by performing a multicentre, retrospective study including patients in 13 referral centers for ALS located in 10 Italian regions, showed that patients with ALS affected by hypertension at diagnosis had a median survival of 37 months as opposed to 49 months for those who were not affected, by concluding that hypertension and heart diseases, as well as hematological diseases, are independently associated with shorter survival [34]. Evidence on the possible impact of cardiovascular drugs on ALS is even more scarce. It is of interest that the riluzole mechanism of action also encompasses sodium and calcium channels modulation; some drugs (e.g., verapamil) identified in our analysis could act oppositely. Beta-blockers could instead worsen respiratory symptoms, by acting as antagonists on bronchial beta-2 receptors.

Role of bisphosphonates

Bisphosphonates are used to treat patients with osteoporosis, to prevent hip and vertebral fractures, and among known side effects, paradoxical bone damage is included: osteonecrosis of the jaw (ONJ) and atypical femoral fracture. Our findings on bisphosphonates as possible risk factors for ALS onset may be due to different reasons. First, patients on bisphosphonates to treat osteoporosis might already have non-diagnosed ALS. Consequently, reporters could have inappropriately identified bisphosphonates as a suspected cause for ALS onset. Bone mineral loss has been noted in ALS and aberrant calcium metabolism and vertebral anomalies have been detected in some patients with ALS [35], and according to the study by Peters T.L. et al poor bone health may be related to ALS [36]. There are in fact some common features

that may link ALS and osteoporosis, including the influence of several neurotoxic metals, such as lead and the age at onset [37].

Again, ALS worsening might depend on paradoxical side effects of bisphosphonates on bone growth, already affected in ALS. In fact, it is likely that skeletal muscle experiences double pathological insults –both intrinsic and extrinsic– during ALS progression, leading to severe atrophy in a very short period of time (3–5 years) after the onset of the ALS symptoms [38].

Signals for drugs used in ALS patients

Among other signals that we detected, several drugs belong to the symptomatic treatment offered to ALS patients in order to control and delay the disease process. We could therefore exclude the following drugs from hypotheses on the biological plausibility of adverse drug effects:

- riluzole and edaravone (both used as treatments of choice for ALS [39]);
- dextromethorphan and quinidine combination (authorized for pseudobulbar syndrome in ALS patients in the US [40]);
- quinine (used to treat cramps [41])
- muscle-relaxant agents (tizanidine, baclofen, and botulinum toxin used to reduce spasticity in ALS patients [42]);
- benzodiazepines (lorazepam, to treat both spasticity and anxiety symptoms [43]);
- immunomodulatory agents (interferon beta-1b, tested in ALS because it might interfere with immune mechanisms involved in the pathogenesis of the disease [44,45]);
- cholecalciferol (offered to contrast neuroinflammation and also because the impact of vitamin D deficiency is considered a favoring factor in various central or peripheral neurological diseases such as ALS [46]);
- antidepressant agents (paroxetine, escitalopram, duloxetine, used in ALS to treat depression, excessive salivation for their anticholinergic side effects, and emotional

outbursts [47,40]). Moreover, as for other neurodegenerative conditions, depression may be a prodromal symptom of ALS. Although motor symptoms are traditionally perceived as the first and main symptoms of ALS, the involvement of nonmotor symptoms both at the onset and after the debut of motor symptoms is likely [47]. This case represents an additional reason for reporting ALS after antidepressant use.

- NSAIDs (used to treat pain, which commonly affects limbs, trunk, and neck in all stages of ALS and becomes more common as the disease progresses [40]).

Nevertheless, it has to be said we cannot totally exclude a role in ALS development for some of the agents listed above. As an example, quinine and quinidine, as well as hydroxychloroquine, might contribute to the worsening of ALS by antagonizing acetylcholine on its nicotinic receptors [48]. At the same time, we cannot exclude that some other promising drug is affected by reverse causality bias. In fact, the ALS presymptomatic stage could last also for years [49], and early non-motor symptoms have been increasingly recognized in the disease [50], increasing the possibility of confounders in this kind of retrospective studies. Thus, we emphasize that the results generated in the present study should be interpreted with caution. Further prospective, population-based, cohort studies may provide more reliable evidence with a lower interference of reverse causation than retrospective studies.

Role of gut microbiota

Finally, ALS development seems to be connected with gut dysbiosis and altered intestinal permeability, although the precise biological mechanisms underlying such a relationship remain to be clarified [8]. It is noteworthy that several classes of drugs with an ALS signal in FAERS are known to alter intestinal microbiota: statins, by interacting with bile acids and impacting upon expression of inflammatory markers that influence microbial community structure; antineoplastic agents, for which a mechanistic framework of interaction with microbiome was recently described (TIMER - Translocation, Immunomodulation,

Metabolism, Enzymatic degradation and Reduced diversity); NSAIDs, by altering the function of the small intestine; antihypertensive drugs, for which animal data suggested interaction with gut microbiota [51,52].

Limitations and Strengths

The analysis of spontaneous reporting data allows for a cheap and timely detection of unexpected drug-event disproportionalities. It collects a great number of reports from a large heterogeneity of environments and accounts for a complexity that is usually excluded from randomized clinical trials (e.g., comorbidity, polytherapy, misuse, extreme ages). Nonetheless, it does not allow formal risk assessment and requires considering multiple biases and limitations. Underreporting is an important problem, particularly for multifactorial and delayed onset events such as ALS is thought to be, therefore spontaneous reporting systems cannot be used to calculate incidence. Furthermore, reports are submitted on a spontaneous basis, both by doctors and consumers, and are often unverified. To account for inter-reporter heterogeneity in the choice of terms, partly due to the redundancy of the MedDRA, we extended the case codification for the broader analysis. We also discussed the reverse causality bias and prioritized disproportionality signals with ROR stability on multiple sensitivity analyses.

5. CONCLUSIONS

With this pharmacovigilance analysis, we detected drugs disproportionally reported with ALS and discussed their potential role. Our findings strengthened the plausibility of a precipitating role of drugs primarily through immunomodulation (e.g., TNF-α, ustekinumab, omalizumab), but also by impacting metabolism and the musculoskeletal integrity (e.g., statins and bisphosphonates). Our findings provide a useful background for planning large cohort studies to reinforce or weaken the plausibility of a causal role for specific drugs in the development of

ALS. They also represent an input for translational research on mechanisms of ALS and on relevant possible therapeutic strategies.

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DECLARATIONS

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Ethics approval: Not applicable

Consent to participate: Not applicable

Consent for publication: Not applicable

Data availability: The data that support the findings of this study were downloaded from the FAERS quarterly data, available to the public at the following link: https://fis.fda.gov/extensions/FPD-QDE-FAERS/FPD-QDE-FAERS.html

Code availability: The R script developed for the analyses is available from the corresponding author upon reasonable request.

Authors contribution: EB, FN, FD, JM, and EP conceived the project, reviewed the manuscript. AG and FM wrote the original draft of the manuscript. AG and FM, performed the investigation, carried out the analyses, and interpreted data. FM performed the investigation, carried out the analyses, interpreted data, and administered the project. EB, LV, FN, JM, and EP contributed to methodological issues, supervised the project, and reviewed the manuscript. FD and EP administered the project. All the authors read and approved the final version.

Tables and Figures legends:

Table 1: Synopsis of disproportionality signals of ALS

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Table 1: Synopsis of disproportionality signals of ALS

Class	Subclass	Drugs ATC code	Disproportionality signals*
ALIMENTARY TRACT AND METABOLISM DRUGS	*VITAMINS*	Colecalciferol A11CC	1 drug with Low ROR stability
BLOOD AND BLOOD FORMING ORGANS DRUGS	*ANTIANEMIC PREPARATIONS*	Mecobalamin B03BA	1 drug with Intermediate ROR stability
CARDIOVASCULAR SYSTEM DRUGS	*CARDIAC THERAPY*	Quinidine C01BA Ubidecarenone C01EB	1 drug with Top ROR stability 1 drug with Intermediate ROR stability
	DIURETICS	Bendroflumethiazide C03AA	1 drug with Low ROR stability
	BETA BLOCKING AGENTS	Sotalol C07AA Atenolol C07AB Bisoprolol C07AB	2 drugs with Top ROR stability 1 drug with Low ROR stability
	CALCIUM CHANNEL BLOCKERS	Verapamil C08DA	1 drug with Intermediate ROR stability
	AGENTS ACTING ON THE RENIN – ANGIOTENSIN SYSTEM	Enalapril C09AA Perindopril C09AA Candesartan C09CA Olmesartan C09CA	1 drug with Top ROR stability 1 drug with High ROR stability 1 drug with Intermediate ROR stability 1 drug with Low ROR stability
	LIPID MODIFYING AGENTS	Atorvastatin C10AA Cerivastatin C10AA Fluvastatin C10AA Lovastatin C10AA Pitavastatin C10AA Pravastatin C10AA Rosuvastatin C10AA Simvastatin C10AA Fenofibrate C10AB Colestyramine C10AC Ezetimibe C10AX	9 drugs with Top ROR stability 1 drug with Intermediate ROR stability 1 drug with Low ROR stability
DERMATOLOGICALS DRUGS	*OTHER DERMATOLOGICALS PREPARATIONS*	Glycopyrronium D11AA	1 drug with Intermediate ROR stability
GENITO-URINARY SYSTEM AND SEX HORMONES DRUGS	*UROLOGICALS*	Dutasteride G04CB	1 drug with Intermediate ROR stability

ANTINEOPLASTIC AND IMMUNOMODULATING AGENTS	*ANTINEOPLASTIC AGENTS*	Bendamustine L01AA Methotrexate L01BA Rituximab L01XC Imatinib L01XE	3 drugs with Top ROR stability 1 drug with low ROR stability
	ENDOCRINE THERAPY	Letrozole L02BG	1 drug with Low ROR stability
	IMMUNOSTIMULANTS	interferon beta-1b L03AB	1 drug with Intermediate ROR stability
	IMMUNOSUPPRESSANTS	Abatacept L04AA Leflunomide L04AA Natalizumab L04AA Tofacitinib L04AA Adalimumab L04AB Etanercept L04AB Infliximab L04AB Tocilizumab L04AC Ustekinumab L04AC	7 drugs with Top ROR stability 1 drug with High ROR stability 1 drug with Intermediate ROR stability
MUSCULO-SKELETAL SYSTEM DRUGS	*ANTINFLAMMATORY AND ANTIRHEUMATIC PRODUCTS*	diclofenac * M01AB celecoxib * M01AH chondroitin M01AX	1 drug with Top ROR stability 1 drug with Intermediate ROR stability 1 drug with Low ROR stability
	MUSCLE RELAXANTS	botulinum toxin M03AX baclofen M03BX tizanidine M03BX	2 drugs with Top ROR stability 1 drug with Intermediate ROR stability
	DRUGS FOR TREATMENT OF BONE DISEASES	alendronic acid M05BA risedronic acid M05BA zoledronic acid M05BA	1 drug with Top ROR stability 1 drug with Intermediate ROR stability 1 drug with Low ROR stability
NERVOUS SYSTEM DRUGS	*ANTIEPILEPTICS*	Phenytoin N03AB	1 drug with Intermediate ROR stability
	PYSCHOLEPTICS	Lorazepam N05BA Melatonin N05CH	2 drugs with Intermediate ROR stability
	PSYCHOANALEPTICS	Escitalopram N06AB Paroxetine N06AB Duloxetine N06AX	1 drug with Top ROR stability 1 drug with High ROR stability 1 drug with Intermediate ROR stability
	OTHER NERVOUS SYSTEM DRUGS	Edaravone N07XX Riluzole N07XX	2 drugs with High ROR stability
ANTIPARASITIC PRODUCTS, INSECTICIDES AND REPELLENTS	*ANTIPROTOZOALS*	Hydroxychloroquine P01BA Quinine P01BC	1 drug with Top ROR stability 1 drug with Intermediate ROR stability
RESPIRATORY SYSTEM DRUGS	*DRUGS FOR OBSTRUCTIVE AIRWAY DISEASES*	Omalizumab R03DX	1 drug with Top ROR stability

	COUGH AND COLD PREPARATIONS	Dextromethorphan R05DA	1 drug with Top ROR stability

*ROR stability: Top ROR stability - signal maintained in 4 out of 4 analyses; High ROR stability - signal maintained in 3 out of 4 analyses; Intermediate ROR stability: signal maintained in 2 out of 4 analyses; Low ROR stability - signal emerged only in 1 out of 4 analysis.

A disproportionality signal was defined when at least 5 cases were submitted, and the lower limit of the ROR 95%CI was higher than 1.