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# Reduction in serum concentration of valproic acid secondary to the intake of ibuprofen as an example of valproic acid auto-induction metabolism

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**Introduction.** In 1998, an unexplained drug-drug interaction between valproic acid (VPA) and ibuprofen was reported. VPA has been considered a moderate inhibitor of several metabolic enzymes, but recently its inductive properties have been described, including the possibility of auto-induction. Ibuprofen can displace VPA from the plasmatic protein, increasing its serum free concentration, and subsequently its pharmacological actions, including auto-induction. The objective of this article is to describe a similar case and to contribute to the clarification of the underlying pharmacokinetic mechanisms.

**Methods.** A 29-year-old Spanish Caucasian male with schizophrenia was followed with steady-state trough serum concentrations of VPA and clozapine for 5 years, including 3 ibuprofen trials. The main outcome variable was the concentration-to-dose (C/D) ratio, a measure of the ability to eliminate a drug. Independent sample Mann-Whitney U tests were performed to compare C/D ratios.

**Results.** Five VPA C/D ratios, contaminated by VPA auto-induction occurring during or shortly after the two latter ibuprofen trials, were significantly lower ($p<0.001$) than the other 34 VPA C/D ratios of VPA not contaminated by auto-induction. During the highest ibuprofen dose in the third trial, the patient had two very low clozapine C/D ratios, which were significantly lower than the other 26 clozapine C/D ratios ($p=0.021$).

**Conclusions.** Reduction in total VPA concentrations could be explained by ibuprofen displacing VPA from the plasma proteins, increasing the serum free VPA. This may induce the metabolism of VPA (and clozapine) and subsequently decrease their serum total concentrations.

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## Reducción en la concentración sérica del ácido valproico secundaria a la toma de ibuprofeno como ejemplo de autoinducción metabólica del ácido valproico

**Introducción.** En 1998, se comunicó una inexplicada interacción farmacológica entre el ácido valproico (AVP) e ibuprofeno. El AVP se ha considerado un inhibidor moderado de varias enzimas metabólicas, pero recientemente se han descrito propiedades inductivas que incluyen la posibilidad de auto-inducción. Ibuprofeno puede desplazar el AVP de las proteínas plasmáticas, aumentando su concentración plasmática libre y consecuentemente sus acciones farmacológicas, incluyendo la auto-inducción. El objetivo de este artículo es describir un caso similar y contribuir a clarificar los mecanismos farmacocinéticos subyacentes.

**Metodología.** En un varón caucásico español de 29 años con esquizofrenia se siguieron durante 5 años las concentraciones plasmáticas de AVP y clozapina, incluyendo 3 ensayos con ibuprofeno. La variable de resultado principal fue la concentración-dosis (C/D) ratio, una medida de la capacidad para eliminar un fármaco. La prueba de U-Mann-Whitney para muestras independientes se utilizó para comparar las C/D ratios.

**Resultados.** Cinco C/D ratios de AVP, contaminadas por auto-inducción de AVP durante o poco después de los dos últimos ensayos con ibuprofeno, fueron significativamente inferiores ($p<0,001$) que las restantes C/D ratios de AVP. Con la mayor de las dosis de ibuprofeno del tercer ensayo, el paciente tuvo dos C/D ratios de clozapina muy bajas que fue-

ron significativamente inferiores a las restantes C/D ratios de clozapina (p=0,021).

**Conclusiones.** La reducción de las concentraciones plasmáticas totales de AVP puede explicarse porque ibuprofeno desplaza el AVP de las proteínas plasmáticas, incrementando el AVP libre. Éste puede inducir el metabolismo del AVP (y de clozapina) y consecuentemente disminuir su concentración plasmática total.

**Palabras clave:** Interacciones farmacológicas, Inducción enzimática, Ibuprofeno, Unión a proteínas, Ácido valproico, Farmacocinética

## INTRODUCTION

Valproic acid (or valproate) was originally approved as an antiepileptic drug (AED) and, more recently, as a mood stabilizer. It is protein-bound, has a half-life of 5-20 hours, and is mainly metabolized in the liver with minimal renal excretion[1]. Valproic acid (VPA), as with many AEDs, has many drug-drug interactions (DDI). Traditionally, VPA was mainly considered an inhibitor[1-4], but more recently has also been considered a potential inducer. The lack of attention to VPA inductive properties is not surprising since the clinical relevance of AED inducers has been systemically undervalued in both epilepsy[5] and bipolar disorder[6]. The potent inductive properties of carbamazepine, phenobarbital and phenytoin are well-established[1-5]. On the other hand, mild AED inducers[5-8] such as oxcarbazepine, topiramate and VPA are clinically relevant inhibitors, but they can also act as clinically relevant inducers in some circumstances. Their mild inductive effects are not as well-established because they: 1) can be obscured by their inhibitory properties, 2) may only be present in high doses, and 3) require even longer (sometimes months) to reach maximum effects or disappear[6,7]. Mild AED inducers may act through the same mechanisms as potent AED inducers, the nuclear receptors, including the constitutive androstane and the pregnane X receptors[8].

Traditionally, VPA was considered to be a moderate inhibitor of several enzymes including the cytochrome P450 (CYP) 2C9 (CYP2C9), the epoxide hydroxylase that metabolizes carbamazepine, and the glucuronidation enzymes that metabolize lamotrigine[1-4,7]. More recently, valproic acid inductive properties have been described[6,7]. First, rat studies suggested that VPA may auto-induce its own glucuronidation[9]. *In vitro* studies described VPA as an inducer of CYP3A4 and P-gp gene expression[10] and vitamin D metabolism[11]. Clinical studies demonstrated that VPA, in some circumstances, has clinically relevant inductive effects on aripiprazole[12], clozapine[13-16], olanzapine[17-19] and irinotecan[20]. A prospective clinical study in volunteers showed that VPA can also induce its own metabolism in humans[21]. This may explain some rare cases of patients needing very high doses of VPA (>4000 mg/day and up to 10,000 mg/day) to keep therapeutic serum VPA concentrations[22].

Smoking induces clozapine metabolism and US male smokers frequently need >600 mg/day of clozapine to reach clozapine therapeutic levels[23,24]. A US African-American male patient who smokes needed 1300 mg/day of clozapine due to further induction by VPA[15]. Surprisingly, 1000 mg/day of VPA did not appear to have any clozapine induction beyond the inductive effects caused by 20 cigarettes per day. However, VPA induction occurred with the addition of 81 mg/day of aspirin, which decreased VPA metabolism and, more importantly, displaced serum VPA from the plasma proteins, contributing to an important increase in free serum VPA concentrations. The free serum VPA fraction explains VPA pharmacological actions, including its own auto-inductive properties, which are going to be higher if the serum free concentrations are higher[15]. This case illustrates the clinical relevance of the VPA-aspirin DDI which has been proposed in prior articles[25-28]. Ibuprofen does not inhibit VPA metabolism but, like aspirin, ibuprofen is known to displace drugs such as VPA from the plasmatic protein, increasing its pharmacological action[28-30].

In 1998, an orthopedic journal[31] reported an interaction between VPA and ibuprofen. A 15-year-old US male taking VPA 700 mg/day for epilepsy was prescribed ibuprofen 2400 mg/day for an ankle fracture. Five days later, the VPA therapeutic drug monitoring (TDM) was subtherapeutic, 43 µg/ml versus 50-100 previously. One week after ibuprofen discontinuation, the serum VPA concentration increased to 60 µg/ml[22]. Since 1998, to our knowledge, this DDI has not been well explained and its clinical relevance and potential deleterious effect is not well-understood. However, it may occur frequently because VPA is commonly prescribed and over-the-counter ibuprofen may be used frequently by VPA patients.

The objective of this article is to report a case of reduction of serum concentration of VPA secondary to the intake of ibuprofen and to propose possible underlying pharmacokinetic mechanisms. We hypothesized that reduced VPA levels could be explained by ibuprofen displacing VPA from the plasma proteins, increasing the serum free VPA. This higher free concentration caused an increase in VPA metabolism and, subsequently, a decrease in serum total VPA concentrations because this patient was particularly sensitive to VPA self-induction[22]. The case includes TDM of VPA and clozapine for 5 years and uses statistics to verify the hypothesis.

## METHODS

### Study and design

This is a single case design. The primary aim of the study was to describe and to analyze the potential pharmacokinetic DDIs in a patient diagnosed with schizophrenia admitted to a day treatment program. The patient and his mother signed an Informed Consent Form for Case Reports whereby they gave permission to use the information included in this case.

### Participant and procedure

The participant is a 29-year-old Spanish Caucasian male whose serum VPA concentrations were unexpectedly found to be subtherapeutic. After a clinical inquiry, the patient explained that he had been taking ibuprofen during the days prior to laboratory testing and that he had quite often taken ibuprofen for several inflammatory processes. A revision of the patient's medical records was carried out; two additional ibuprofen trials were identified in five years of records.

#### *Psychiatric History*

The patient started having psychiatric symptoms at the age of 17 when he failed to achieve the expected level of educational/occupational functioning, but was not seen by a psychiatrist until age 20 when he had trouble completing a software training course. He was diagnosed with DSM-IV schizophrenia, disorganized type, and received, successively, treatment with risperidone up to 9 mg, olanzapine up to 30 mg and paliperidone up to 15 mg with little improvement. At age 25, after isolating himself at home for over 15 months, he was involuntarily admitted to a psychiatric hospital and then was referred to the day treatment program.

At admission, he presented distractibility and hyper-vigilance, bizarre behavior (severe stereotypes and manneriisms), spontaneous speech characterized by poverty in the amount and the content, verbal stereotypes, poverty of thought and thought blocking, blunted affect, auditory hallucinations and, possibly, persecutory delusions, which he refused to talk about. In addition to the rehabilitative program, he received pharmacological treatment including a combination of clozapine 600 mg/day, long-acting risperidone 50 mg IM every 2 weeks, and VPA oral solution 800 mg/day, which brought about a substantial improvement; he was able to apply for a transitional job. He was discharged from the day program and followed by an outpatient psychiatrist for 3 years who changed the long-acting risperidone to paliperidone palmitate 150 mg IM every 28 days. During the second year of follow-up, after several stressful life events, he suffered a partial psychotic relapse and was referred for the second time to the day treatment program. Risperidone 3 mg/day was added and his symptoms gradually diminished.

#### *Ibuprofen trials*

After 4 months during this second stay at the day program, serum VPA concentrations were unexpectedly found to be subtherapeutic (14.1 µg/ml), even though the patient and his mother insisted that the patient had been taking the VPA oral solution regularly. Plasma clozapine concentrations were slightly lower (358 ng/ml) than previous levels. The patient explained that for the 11 days prior to the laboratory test, he had been taking ibuprofen 1800 mg/day for bronchitis. Eleven days after ibuprofen was discontinued, VPA levels increased to 39.2 µg/ml and 6 weeks after ibuprofen discontinuation returned to a therapeutic range (53.9 µg/ml).

Two additional ibuprofen trials were identified in the records (Table 1). The first trial included a 5-day treatment with 1200 mg/day during a prior bronchitis illness. No VPA concentration was measured that month, but the one during the next month (3 weeks after ibuprofen discontinuation) was slightly lower (36.0 µg/ml versus 40s-50s in prior months). The second trial of 1200 mg/day lasted 2 months, was started to treat bronchitis and then continued for an ankle sprain. This second ibuprofen trial was associated with two subtherapeutic VPA concentrations, 24 and 23 µg/ml, respectively.

### Outcome variables and information sources

The main outcome variables (Table 1) were the steady-state trough concentrations of clozapine and VPA measured in commercial laboratories, the clozapine concentration-to-dose ratios (C/D ratios), the VPA C/D ratios and the VPA C/D ratios multiplied by 1000. Moreover, the authors' agreement regarding the drug interaction was evaluated by the Drug Interaction Probability Scale (DIPS)[32].

#### *Concentration-to-dose (C/D ratios)*

The C/D ratio is used by pharmacologists as a measure of the ability to eliminate a drug. The first published article using the C/D ratio demonstrated the effects of inducers and inhibitors on clozapine metabolism[33]. The C/D ratio has increasingly been used for personalizing the dosing of

**Table 1** Valproic acid, ibuprofen and clozapine doses and steady-state trough concentrations

| Days | Valproic acid | | | | Ibuprofen | Clozapine | | |
|---|---|---|---|---|---|---|---|---|
| | D (mg/day) | C (µg/ml) | C/D | C/D x 1000 | D (mg/day) | D (mg/día) | C (ng/ml) | C/D |
| | | | | PSYCHIATRIC ADMISSION | | | | |
| 1[a] | 200 | | | | 0 | 0 | | |
| 4[a] | 400 | | | | 0 | 0 | | |
| 16[a] | 400 | 52.4 | 0.131 | 131 | 0 | 50 | | |
| 37[b] | | | | DAY TREATMENT PROGRAM (First stay) | | | | |
| 40[b] | 600 | 47.7 | 0.080 | 80 | 0 | 300 | | |
| 54[b] | 600 | 48.6 | 0.081 | 81 | 0 | 300 | 304 | 1.01 |
| 75[b] | 800 | 48.6 | 0.061 | 61 | 0 | 400 | 339 | 0.85 |
| 102[c] | 800 | 63.0 | 0.079 | 79 | 0 | 400 | 262 | 0.66 |
| 187[d] | 800 | 50.7 | 0.063 | 63 | 0 | 600 | 408 | 0.68 |
| 207[e] | 800 | 51.0 | 0.064 | 64 | 0 | 600 | 412 | 0.69 |
| | | | | OUTPATIENT FOLLOW-UP | | | | |
| 354[e] | 800 | 45.6 | 0.057 | 57 | 0 | 600 | | |
| 383[e] | 800 | 41.7 | 0.052 | 52 | 0 | 600 | | |
| 414[f] | 800 | 51.0 | 0.064 | 64 | 0 | 600 | | |
| 445[f] | 800 | 50.6 | 0.063 | 63 | 0 | 600 | | |
| 474[f] | 800 | 53.1 | 0.066 | 66 | 0 | 600 | | |
| 505[f] | 800 | 46.2 | 0.058 | 58 | 0 | 600 | | |
| 535[f] | 800 | 49.3 | 0.062 | 62 | 0 | 600 | | |
| 565[f] | 800 | 49.8 | 0.062 | 62 | 0 | 600 | | |
| 596[f] | 800 | 52.7 | 0.066 | 66 | 0 | 600 | | |
| 624[f] | 800 | 54.4 | 0.068 | 68 | 0 | 600 | | |
| 660[f] | 800 | 50.8 | 0.064 | 64 | 0 | 600 | | |
| 695[f] | 800 | 45.7 | 0.057 | 57 | 0 | 600 | 474 | 0.79 |
| 726[f] | 800 | 43.9 | 0.055 | 55 | 0 | 600 | 555 | 0.93 |
| 745[g] | 800 | | | | 1200 | 600 | | |
| 750[g] | 800 | | | | 1200 | 600 | | |
| 751[f] | 800 | | | | 0 | 600 | | |
| 772[f] | 800 | 36.0 | 0.045 | 45 | 0 | 600 | 490 | 0.82 |
| 803[h] | 800 | 55.2 | 0.069 | 69 | 0 | 600 | 630 | 1.05 |
| 826[h] | 800 | 46.0 | 0.058 | 58 | 0 | 600 | 447 | 0.75 |
| 862[h] | 800 | 48.8 | 0.061 | 61 | 0 | 600 | 512 | 0.85 |
| 892[h] | 800 | 52.7 | 0.066 | 66 | 0 | 600 | 607 | 1.01 |
| 922[h] | 800 | 45.0 | 0.056 | 56 | 0 | 600 | 406 | 0.68 |
| 953[h] | 800 | 46.1 | 0.058 | 58 | 0 | 600 | 580 | 0.97 |
| 984[h] | 800 | 45.1 | 0.056 | 56 | 0 | 600 | 602 | 1.00 |
| 1019[h] | 800 | 40.3 | 0.050 | 50 | 0 | 600 | 534 | 0.89 |
| 1032[h] | 800 | | | | 1200 | 600 | | |
| 1046[h] | 800 | 24.2 | 0.030 | 30[m] | 1200 | 600 | 563 | 0.94 |

**Table 1** Continuation

| Days | Valproic acid | | | | Ibuprofen | Clozapine | | |
|---|---|---|---|---|---|---|---|---|
| | D (mg/day) | C (µg/ml) | C/D | C/D x 1000 | D (mg/day) | D (mg/día) | C (ng/ml) | C/D |
| | | | | OUTPATIENT FOLLOW-UP | | | | |
| 1083[h] | 800 | 23.3 | 0.029 | 29[m] | 1200 | 600 | 552 | 0.92 |
| 1086[h] | 800 | | | | 1200 | 600 | | |
| 1087[h] | 800 | | | | 0 | 600 | | |
| 1138[h] | 800 | 57.1 | 0.071 | 71 | 0 | 600 | 544 | 0.91 |
| 1195[h] | 800 | 62.0 | 0.078 | 78 | 0 | 600 | 701 | 1.17 |
| 1227[h] | 800 | 54.3 | 0.068 | 68 | 0 | 600 | 412 | 0.69 |
| 1257[h] | 800 | 51.1 | 0.064 | 64 | 0 | 600 | 321 | 0.54 |
| 1285[i] | | | | DAY TREATMENT PROGRAM (Second stay) | | | | |
| 1290[j] | 800 | 54.5 | 0.068 | 68 | 0 | 600 | 501 | 0.84 |
| 1400[j] | 800 | | | | 1800 | 600 | | |
| 1411[k] | 800 | 14.1 | 0.018 | 18[m] | 1800 | 600 | 358 | 0.60[l] |
| 1414[k] | 800 | | | | 1800 | 600 | | |
| 1415[k] | 800 | | | | 0 | 600 | | |
| 1425[k] | 800 | 39.2 | 0.049 | 49[m] | 0 | 600 | | |
| 1439[k] | 800 | 38.9 | 0.049 | 49[m] | 0 | 600 | 371 | 0.62[l] |
| 1467[k] | 800 | 53.9 | 0.067 | 67 | 0 | 600 | 435 | 0.73 |
| 1534[k] | 800 | 46.2 | 0.058 | 58 | 0 | 600 | 487 | 0.81 |
| 1562[k] | 800 | 45.8 | 0.057 | 57 | 0 | 600 | 586 | 0.98 |

D: Dose; C: serum concentration; C/D ratio: concentration-to-dose ratio.
[a]Other medications included risperidone 6 mg/day, risperidone long-acting 100 mg every 14 days, and pravastatin 20 mg/day.
[b]Other medications included risperidone long-acting 100 mg every 14 days, and pravastatin 20 mg/day.
[c]Other medications included risperidone long-acting 100 mg every 14 days and simvastatin 20 mg/day.
[d]Other medications included risperidone long-acting 75 mg every 14 days and simvastatin 20 mg/day.
[e]Other medications included risperidone long-acting 50 mg every 14 days and simvastatin 20 mg/day.
[f]Other medications included risperidone long-acting 50 mg every 14 days and atorvastatin 40 mg/day.
[g]Other medications included risperidone long-acting 50 mg every 14 days, atorvastatin 40 mg/day, and from 12/12/12 to 12/20/12 amoxicillin, 1500 mg/day .
[h]Other medications included risperidone long-acting 50 mg every 14 days, atorvastatin 40 mg/day and fenofibrate 200 mg/day.
[i]Other medications included paliperidone palmitate 150 mg every 28 days, atorvastatin 40 mg/day and fenofibrate 200 mg/day.
[j]Other medications included paliperidone palmitate 150 mg every 28 days, risperidone 3 mg/day, atorvastatin 40 mg/day, fenofibrate 200 mg/day and amoxicillin 2400 mg/day (7 days)
[k]Other medications included paliperidone palmitate 150 mg every 28 days, risperidone 3 mg/day, atorvastatin 40 mg/day and fenofibrate 200 mg/day.
[l]An independent sample Mann-Whitney U test provided a significant difference (p=0.021) when comparing these 2 clozapine C/D ratios with the other 26 clozapine C/D ratios (median 0.85). These 2 clozapine C/D ratios during the third ibuprofen trial were presumably associated with an increased free serum VPA concentration and subsequent clozapine induction, while the other 26 clozapine C/D ratios reflect periods with no clozapine induction.
[m]An independent sample Mann-Whitney U test provided a very significant difference (p<0.001) between these 5 VPA C/D ratios (median 30) and the other 34 VPA C/D ratios on 800 mg/day (median 63). These 5 VPA C/D ratios were presumably contaminated by VPA auto-induction secondary to increased VPA free concentrations due to ibuprofen treatments, while the other 34 VPA C/D ratios were not influenced by VPA auto-induction.

clozapine[23], risperidone[23], clobazam[34] and, more recently, VPA[22]. The C/D ratio is influenced by genetic, personal and environmental factors. Inducers decrease the C/D ratio and inhibitors increase the C/D ratio. In comparing individuals taking the same drug, a very low C/D ratio indicates an individual with very fast metabolism, while a very high C/D ratio indicates one with very slow metabolism[23].

Clozapine, like the majority of drugs used in neuropsychopharmacology, follows linear kinetics which means that

a linear relationship exists between doses and serum concentrations. The relationship between concentration and dose is stable; it does not change with different doses and concentrations. The drug C/D ratio is constant in the same patient as long as there are no changes in environmental or personal variables[23]. In the US, clozapine C/D ratios, obtained by dividing the serum clozapine concentration (ng/ml) by the clozapine daily dosage (mg/day), typically range between 0.6 and 1.2[23]. US male smokers usually have a concentration ≥350 ng/ml with a dose of 600 mg/day; the C/D ratio is 0.58 (350/600). US female non-smokers usually have a concentration ≥350 ng/ml with a dose of 300 mg/day; the C/D ratio is 1.17 (350/300)[23].

The relationship between VPA dose and total concentration is non-linear; the concentration does not increase proportionally with the dose but increases to a lesser extent due to saturable plasma-protein binding[22,35]. As the VPA C/D ratio is not constant and changes with different doses and concentrations, one must further interpret VPA C/D ratios in the context of a set of concentrations, or a set of doses. To more easily understand the VPA C/D ratio, these values can be multiplied by 1000. If a VPA dose of 2000 mg/day provides a total VPA concentration of 100 µg/mL, the C/D ratio would be 0.050 (100/2000), and the C/D ratio multiplied by 1000 would be 50[22].

## Statistical analysis

Independent sample Mann-Whitney U tests (Table 1) were used to compare clozapine or VPA C/D ratios presumably associated with an increased free serum VPA concentration with the other clozapine or VPA C/D ratios. The statistical analysis was performed with the SPSS® statistics program (version 21.0; IBM-SPSS, Chicago, IL; USA)

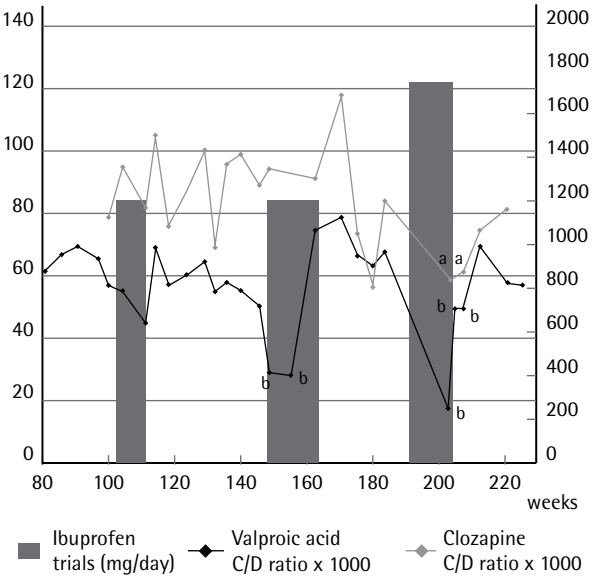
# RESULTS

## Therapeutic drug monitoring

Table 1 describes dosages and steady-state trough serum concentrations of clozapine and VPA during the last 5 years. Concomitant factors include smoking 20-25 cigarettes/day and drinking caffeine 280-440 mg/day (2-3 cups of caffeinated coffee and 1-2 caffeinated colas/day). The patient did not consume alcohol or illegal drugs. Ibuprofen TDM is rarely used and was not available.

## Clozapine C/D ratios

This patient had 28 clozapine measures with a mean ± standard deviation (SD) C/D ratio of 0.83 (±0.16), which is normal for a Caucasian smoker with substantial caffeine intake. During the highest ibuprofen dose in the third trial, the patient had two very low clozapine C/D ratios, 0.60 and 0.62, which were significantly lower than the other 26 clozapine C/D ratios (p=0.021; Table 1, footnote I; Figure 1).




[a]An independent sample Mann-Whitney U test provided a significant difference (p=0.021) when comparing these 2 clozapine C/D ratios with the other 26 clozapine C/D ratios (median 0.85). These 2 clozapine C/D ratios during the third ibuprofen trial were presumably associated with an increased free serum VPA concentration and subsequent clozapine induction, while the other 26 clozapine C/D ratios reflect periods with no clozapine induction.

[b]An independent sample Mann-Whitney U test provided a very significant difference (p<0.001) between these 5 VPA C/D ratios (median 30) and the other 34 VPA C/D ratios on 800 mg/day (median 63). These 5 VPA C/D ratios were presumably contaminated by VPA auto-induction secondary to increased VPA free concentrations due to ibuprofen treatments, while the other 34 VPA C/D ratios were not influenced by VPA auto-induction.

*Figure 1* | *Valproic acid and clozapine C/D ratios and ibuprofen trials*

## VPA C/D ratios

With a VPA dose of 800 mg/day, this patient had 39 serum VPA concentrations. The mean ± SD VPA C/D ratio multiplied by 1000 would be 59±12.1. The second ibuprofen trial with 1200 mg/day over 47 days appeared to be

associated with a decrease in VPA C/D ratio; multiplied by 1000, values would be 20 and 29 during the middle of the ibuprofen trial. The third ibuprofen trial with 1800 mg/day over the course of 15 days was associated with a decrease to 19 in the VPA C/D ratio (multiplied by 1000). The VPA C/D ratios, multiplied by 1000, were 49 and 49, 11 and 25 days after ibuprofen discontinuation, reflecting the residual effects of induction. These 5 VPA C/D ratios (median 30 when multiplied by 1000), contaminated by VPA auto-induction occurring during or shortly after ibuprofen trials, were significantly lower ($p<0.001$; Table 1, footnote m; Figure 1) than the other 34 VPA C/D ratios on 800 mg/day of VPA (median 63) not contaminated by auto-induction.

In the experience of the senior author, in only a few patients[22] does VPA act as an inducer of its own metabolism. These patients are identified by the need to progressively increase the VPA dose to keep the serum total VPA concentration within the therapeutic range. The first VPA C/D ratio multiplied by 1000 on 400 mg/day of VPA was extremely high, 131, compared with the rest of the 5-year period, but it may be due to non-linear kinetics. The first VPA C/D ratios multiplied by 1000 on 600 mg/day, were high, 80 and 81. Once the patient was stable on 800 mg/day of VPA, he never reached a C/D ratio multiplied by 1000 of 80. These two VPA C/D ratios on 600 mg/day may be compatible with VPA auto-induction.

## DIPS

All the authors agreed that the DIPS[32] provided a score of 10 for the effect of ibuprofen leading to VPA auto-induction, which corresponds to a highly probable DDI in this case (scored as 1 point each for questions 1,2, 3, 4, 5, 7, 8 and 10, and 2 points for question 6). All the authors agreed that the DIPS[32] provided a score of 7 for the effect of ibuprofen on VPA leading to clozapine induction, which corresponds to a probable DDI (scored as 1 point each for questions 1, 2, 3, 4, 5, 7 and 8).

## DISCUSSION

### Pharmacological explanation

This 29-year-old Spanish Caucasian male was followed with VPA and clozapine TDM for 5 years during which he had 3 ibuprofen trials. Ibuprofen is known to displace drugs such as VPA from the plasmatic protein, increasing their pharmacological action[28,29]. This case of reduced serum VPA concentrations is explained by ibuprofen displacing VPA from the plasma proteins, increasing the serum free VPA. This higher free concentration caused an increase in VPA metabolism and, subsequently, a decrease in serum total VPA concentrations. There were no VPA concentrations during the first ibuprofen trial. The VPA C/D ratios were significantly lower ($p<0.001$) and compatible with VPA auto-induction during the second and third ibuprofen trials in a patient prone to VPA auto-induction. The limited clozapine levels were also compatible with the ibuprofen-induced clozapine metabolism demonstrated by a significant ($p=0.021$) decrease in clozapine C/D ratios during the third ibuprofen trial. On the other hand, ibuprofen TDM is rarely used and was not available. Ibuprofen is thought to be metabolized by CYP2C8 and CYP2C9[36]. As VPA is a CYP2C9 inhibitor, it is possible that this patient may have had decreased ability to metabolize ibuprofen that may have contributed to increased effects of ibuprofen on VPA protein binding.

### Limitations

We need to acknowledge that DDI associated with protein binding displacement is a controversial issue and very few clinical cases have been published. Literature reviews in the 1970s and 1980s[37] suggested that protein binding should be relevant for DDI. More recent literature suggests that protein binding is rarely relevant for DDI[38] or is generally of minimal clinical significance, but this assumption lacks evidence[29]. The progressive accumulation of information regarding the DDI between aspirin and VPA[15,25-28,30] and between ibuprofen and VPA, including the prior case[31] and this case, further demonstrates the clinical relevance of a DDI involving displacement from protein binding.

Unfortunately, free VPA TDM was not available. We would expect that these levels should have been elevated during the ibuprofen trials. Elevated free VPA concentrations may increase VPA metabolism in the short term but will not explains that it took several weeks to normalize VPA and clozapine metabolism after ibuprofen discontinuation. We think that the elevation of free VPA secondarily increases VPA inductive properties, which had taken several weeks to disappear. This would explain the delay in the normalization of VPA and clozapine and metabolism in some of the ibuprofen trials.

As with other case reports, this was a challenging case. The pharmacological data is somewhat limited since the patient was treated with polypharmacy and went through complex changes in medications, but at least we had comprehensive TDM for 5 years. Likewise, during the third ibuprofen trial the patient was treated in a day hospital that offers a 6-8 hours/day treatment plan Monday through

Friday. This allowed close follow-up and an accurate evaluation of polypharmacy, as well as more precise control of the use of tobacco, caffeine, alcohol and other substances. Although the patient and his mother insisted that the patient had been taking the VPA and the other psychotropic drugs regularly, we cannot be sure about this point, but the pharmacokinetic profile suggests that induction, more than lack of compliance, explains these decreases in VPA and clozapine TDM. During the third ibuprofen trial, although serum VPA levels were found to be subtherapeutic (14.1 µg/ml), plasma clozapine levels were only slightly lower (358 ng/ml) than previous ones, but within the therapeutic range. This was compatible with appropriate compliance and greater induction of VPA metabolism than of clozapine metabolism. As a matter of fact, the analysis of the three ibuprofen trials suggests that, in this patient, adding ibuprofen was associated with greater and more consistent diminutions of VPA C/D ratio than of clozapine C/D ratio.

## CONCLUSIONS

During and shortly after the second and third ibuprofen trials, VPA C/D ratios were significantly lower ($p<0.001$) and compatible with VPA auto-induction in a patient prone to VPA auto-induction. The more limited clozapine levels were also compatible with the ibuprofen-induced clozapine metabolism demonstrated by a significant ($p=0.021$) decrease in clozapine C/D ratios during the third ibuprofen trial. Ibuprofen, by increasing the serum free VPA concentrations, caused a temporal induction of VPA metabolism and possibly of clozapine metabolism, too. Although this patient had no apparent clinical complications from this DDI, the potential interference with the mood stabilizer regimen might have led to a psychotic relapse. Obviously, in the case of a patient with poorly controlled bipolar disorder or epilepsy, the risk of relapse would be higher. Specific pharmacokinetic studies of this DDI are necessary to prevent potential risks to patients.

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### CONFLICT OF INTERESTS

No commercial organizations had any role in the completion or publication of this study. This article was completed without any external funding. In the last 36 months, Dr. Lana has received funds from Janssen-Cilag (2 times) and Otsuka (2 times) to pay for registration at scientific meetings, travel funds from Janssen-Cilag (1 time) and Otsuka (1 time) for scientific meetings, and honoraria from Janssen-Cilag (3 times) for a CME presentation developed by him. In the last 36 months, Dr. Martí-Bonany has received travel funds from Pfizer, Janssen-Cilag, Otsuka and Esteve to attend scientific meetings, an educational grant from Otsuka to pay for a university online course and payment as a blind investigator in a Janssen study. In the last 3 years, Dr. Fuster has received funds from Janssen-Cilag (2 times) and Otsuka (1 time) to pay for registration at scientific meetings. Dr. de Leon declares no competing interest during the last 36 months.

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