Adverse Effects of Etoricoxib, a Selectively Inhibiting Cyclo-Oxygenase-2 Anti-Inflammatory Drug, Assessed on Ants Used as a Model

Marie-Claire Cammaerts1* and Roger Cammaerts2

1Independent Researcher, Retired from the Biology of Organisms Department, University of Brussels, Belgium
2Independent Researcher, Retired from the Natural and Agricultural Environmental Studies Department (DEMNA) of the Walloon Region, Belgium

*Corresponding Author: Marie-Claire Cammaerts, Independent Researcher; Retired from the Biology of Organisms Department, University of Brussels, Belgium.

Received: March 22, 2019; Published: May 02, 2019

Abstract

We examined on ants used as a model the potential adverse effects of etoricoxib, an anti-inflammatory drug of the new generation, inhibiting specifically the COX2 enzyme. We found that this drug decreased the ants’ food consumption, activity, cognition, escaping ability, conditioning, short-term and middle-term memory. The drug increased the ants’ linear speed and sinuosity of movement and tactile perception, and did not impact their orientation capability, audacity and social relationships. No adaptation occurred to these effects, ants developed no dependence on etoricoxib consumption, and after weaning the effect of the drug vanished slowly and linearly, in a total of about 21 hours. On basis of these findings and on those previously obtained on ants for diclofenac and meloxicam, two traditional anti-inflammatory drugs, etoricoxib appears to present less adverse effects than diclofenac. Compared to etoricoxib and diclofenac, meloxicam appears not to change the locomotion and memorization abilities. Instructions for use and scientific literature also suggest less harmful effects using meloxicam instead of etoricoxib or diclofenac.

Keywords: Cognition; Dependence; Food Consumption; Myrmica sabuleti; Sensitivity

Abbreviations

ang.deg.: Angular Degrees; ang.deg./cm: Angular Degrees Per cm; mm/s: Millimeter Per Second; χ2: Chi-Square; vs: Versus; n°: Number; cm: Centimeter; mm: Millimeter; ml: Milliliter; mg: Milligram; s: Second; min: Minute; h: Hours; t: Time; %: Percentage

Introduction

Anti-inflammatory drugs are among the most used medicines for humans as well as for animals. This results from the fact that any inflammatory reaction is painful, and that it causes several unsafe secondary effects such as blood coagulation problems [1], diseases in organs [2], thrombosis [3] and psychiatric perturbations [4].

Nonsteroidal anti-inflammatory drugs (NSAIDs) inhibit two isoforms of the cyclooxygenase (COX) enzyme, COX-1 and COX-2. The constitutive enzyme COX-1 is involved in homeostatic functions such as gastric cytoprotection and platelet aggregation while the inducible enzyme COX-2 activates among others inflammation reaction, pain and fever [5]. Traditional NSAIDs such as diclofenac and meloxicam inhibit COX-1 as well as COX-2 at an almost similar extent [6,7], what can lead to serious gastrointestinal (GI) and blood impairments [8,9]. Therefore, a new generation of much more selective COX-2 NSAIDs, the ‘coxibs’, (celecoxib, rofecoxib, valdecoxib, etoricoxib and...
lumiracoxib) has been produced, deemed to have comparable efficacy as the traditional NSAIDs with a better GI safety profile [10,11]. For instance, the use of etoricoxib was shown to induce fewer GI uncomplicated adverse events than that of diclofenac [12,13]. Coxibs were thus progressively preferred to the traditional NSAIDs, but gave rise to an increased concern about the elevated risks of cardiovascular (CV) events. For some of them, such as rofecoxib, a definitive evidence led to their withdrawal [7,11]. Compared to the use of diclofenac, that of etoricoxib did not contribute to higher cardiovascular risks when experimented in randomized trials [12]. However, only a comparison with a placebo would enable to establish the true risk [7]. Using the precautionary principle, many countries only make celecoxib and etoricoxib available on their pharmaceutical market [11]. Indeed, a collaborative meta-analysis [14] showed that coxibs and diclofenac increase the risk of major cardiovascular events by 75% and 70% in comparison to a placebo, while serious upper GI complications significantly increased too. Another data analysis on the effects of celecoxib and etoricoxib on chronic inflammatory bowel disease [15], however on small samples and short follow-up durations, could not find evidence for cardiovascular or renal toxicity, but the authors were aware that further randomized controlled trials are necessary for defining the safety of these coxibs.

We here intend to examine the ethological and physiological harmful impacts of one largely used 'coxib', etoricoxib, on ants used as models, exactly as we examined such impacts of diclofenac and meloxicam, two traditional anti-inflammatory drugs [16,17].

Here below, we give some information on the here studied drug; we explain why we used ants, which species we employed, and what we know on it; and we list the physiological and ethological traits we examined. Then, after having briefly related our methods and given our results, we compare them to those of the literature and conclude.

Etoricoxib (Figure 1), a sulfone coxib the effects of which are here examined, is the active component of the drug ‘arcoxia®’ (MSD, Netherlands) available in Europe. In the information for use joined to the package, it is mentioned that consuming this drug may induce some adverse effects such as stomach pain, digestive problems, oedema, heart problems, allergy, problems of appetite, and anxiousness. No studies could be found as for the impact of etoricoxib on several patients’ important physiological and ethological traits such as the activity, audacity, cognition, memory, social relationships, adaptation to the adverse effects of the drug, habituation to its beneficial effects, and dependence on its consumption. Moreover, the already made studies were performed on patients presenting some inflammatory reaction, what makes impossible detecting the effects due only to the drug. Therefore, studying the effects of etoricoxib on animals (i.e. ants) not suffering from inflammation and looking to the traits here above mentioned, could be useful.

Why do we used ants as models?

Animals’ and humans’ physiological and ethological traits are fundamentally similar [18]. This is why they are firstly examined on animals as models (e.g. fruit flies, cockroaches, bees, mice, monkeys) then on humans [19]. Invertebrates are advantageously used as models due to their rapid development and easy maintenance in a laboratory [20]. Insects, hymenoptera among others, are often used [21] and

Citation: Marie-Claire Cammaerts and Roger Cammaerts. “Adverse Effects of Etoricoxib, a Selectively Inhibiting Cyclo-Oxygenase-2 Anti-Inflammatory Drug, Assessed on Ants Used as a Model”. *EC Pharmacology and Toxicology* 7.5 (2019): 373-392.
Adverse Effects of Etoricoxib, a Selectively Inhibiting Cyclo-Oxygenase-2 Anti-Inflammatory Drug, Assessed on Ants Used as a Model

ants can thus also be used [22]. These insects are eu-social, with colonial regulation, labor division and exchange of information thanks to tactile and chemical signals (pheromones) [23-25]. The ants take care of their brood, construct sophisticated nests, and chemically mark the different parts of their territory [23]. They navigate, recruit congeners, relocate their nest, clean the inside of it, and create cemeteries [24]. According to their complex biology, they can serve as biological models. The impact of substances and environmental changes can be studied on them, and hypothesis about the impact of these factors on other organisms including humans can be emitted.

Which species did we use?

We have largely studied some species of the genus Myrmica, having looked, among others, to their ecology, eyes morphology, angle of vision, visual perception, recruitment strategies, navigation systems, conditioning [26], to the ontogenesis of some of their abilities [27], and to the limit of their cognitive abilities [28]. Studying on them the effect of manmade electromagnetism revealed that they could be good biological models [29,30]. Effectively, they were so all along our studies of the impact of products used by humans [31-33]. Recently, we have used again the ant M. sabuleti Meinert 1861 as a model for studying the effect of diclofenac and meloxicam on several physiological and ethological traits [16,17]. Here, we used once more M. sabuleti for examining in the same way the physiological and ethological potential impacts of one of the newly produced anti-inflammatory drugs.

Which traits did we examine?

First on ants under normal diet (controls), then on the same ants consuming etoricoxib (tests), we examined 19 traits: meat consumption, sugar water consumption, general activity, speed of locomotion, sinuosity of movement, orientation ability, audacity, tactile perception, brood caring, aggressiveness against nestmates, aggressiveness against aliens, cognition, escaping ability, conditioning capability and short term memory, adaptation to adverse effects of the drug (if any), habituation to beneficial effects of the drug (if any), dependence on the drug consumption, and decrease of the effect of the drug (if any) after weaning.

Adaptation to a product occurs when adverse effects caused by that product decrease over time. Habituation to a product occurs when beneficial effects caused by that product decrease over time. Dependence on a product develops when individuals using this product prefer a life including this product than a life without it.

Materials and Methods

Collection and maintenance of ants

The experimental work was conducted on two colonies of M. sabuleti (colonies A and B) collected, in September 2018, in an abandoned quarry of the Aise valley (Ardenne, Belgium). A third colony, collected the same day in that valley, was used to perform the control experiment of the conditioning study. A fourth colony, collected in June 2018 at Marchin (Condroz, Belgium), also in an abandoned quarry, provided the alien ants used for studying the ants’ aggressiveness. Each colony contained about 500 workers, brood and a queen. They were maintained in the laboratory in two to three glass tubes half filled with water, a cotton plug separating the ants from the water. The nest tubes of each colony were set in a tray (34 cm x 23 cm x 4 cm) serving as foraging area. In these trays, pieces of Tenebrio molitor larvae (Linnaeus, 1758) were deposited three times per week, and cotton plugged tubes filled with sugar water were permanently set. The ambient temperature was ca 20°C, the humidity 80%, the lighting 330 lux while working on ants, and the electromagnetism 2 µWm², optimum environmental conditions for the species. The ants of a same colony are here often named ‘nestmates’ as researchers on social insects commonly do.

Solution of etoricoxib given to the ants

We used the drug labeled ‘arcoxia®’ (MSD-Chibret) the active substance of which is etoricoxib. Information on arcoxia® available on internet and in the instructions for use supplied with this drug advice patients to consume 30 mg, 60 mg, 90 mg or 120 mg per day according to the kind and the intensity of the inflammation. We opted for the middle dose of 60 mg per day. Tablets containing this dose
were furnished by the pharmacist Wera (1170 Bruxelles). A human commonly consume one liter of water each day. Consequently, humans treated with 60 mg of arcoxia® per day consume this together with one liter of water. Insects, and thus ants, need about 10 times less water than mammals, due to their different skin, respiratory apparatus and excretory system [18]. Therefore, for being under an etoricoxib diet equivalent to that of a human treated with 60 mg of the drug ‘arcoxia®’ per day, the ants must be provided with a concentration of 60 mg of this drug in 100 ml of water (or any liquid). We gave thus to the ants such a concentration using their usual sugar water as liquid and delivered this solution in their usual tubes plugged with cotton. The ants could drink this solution ad libitum, according to their size and request. We checked many times per day that they effectively did so.

**Assessment of the examined traits**

These assessments have been many times explained in previous studies [31-33] and are thus only briefly related here. Each trait was firstly assessed on ants under normal diet, then on the same ants consuming etoricoxib, except adaptation, dependence and loss of the effects which were assessed only on ants consuming or having consumed etoricoxib.

**Food consumption, general activity**

While ants were under normal diet then when they consumed etoricoxib, we counted during six days, six times per day at the same times o’clock each day, the ants eating the pieces of *T. molitor* larvae, those drinking the sugar water, and those being active at any place of their environment [31-33]. The mean of the daily counts was established for each kind of count (Table 1) and the six obtained means were used for statistical analysis. For information only, for each kind of count, the average of the six means was calculated (Table 1, last line).

<table>
<thead>
<tr>
<th>Experimental days</th>
<th>Normal diet</th>
<th>Diet with etoricoxib</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Meat</td>
<td>Sugar water</td>
</tr>
<tr>
<td>I</td>
<td>1.67</td>
<td>2.00</td>
</tr>
<tr>
<td>II</td>
<td>2.00</td>
<td>2.83</td>
</tr>
<tr>
<td>III</td>
<td>1.50</td>
<td>3.67</td>
</tr>
<tr>
<td>IV</td>
<td>1.67</td>
<td>3.00</td>
</tr>
<tr>
<td>V</td>
<td>2.67</td>
<td>2.83</td>
</tr>
<tr>
<td>VI</td>
<td>1.67</td>
<td>2.17</td>
</tr>
<tr>
<td>I - VI</td>
<td>1.86</td>
<td>2.75</td>
</tr>
</tbody>
</table>

*Table 1: Impact of etoricoxib on food consumption and general activity. Mean of daily counts (lines I to VI) and of the six daily means (last line: I - VI). Under etoricoxib diet, the ants consumed less meat and sugar water, and were less active.*

**Linear and angular speeds, orientation**

These traits were assessed on ants moving in their foraging area, the linear and angular speeds without stimulating the ants, the orientation by stimulating them with a nestmate tied to a piece of paper (Figure 2A). Such a nestmate emits its mandibular glands attractive alarm pheromone [31-33]. For the speeds on one hand and for the orientation on the other hand, 40 ants’ trajectories were recorded, and were then analyzed thanks to appropriate software [34] based on the three following definitions. The linear speed (in mm/s) is the length of a trajectory divided by the time spent to travel it; the angular speed (in angular degrees/cm = ang.deg./cm) is the sum of the angles made by successive adjacent segments, divided by the length of the trajectory; the orientation (in ang. deg.) towards a location is the sum of the successive angles made by the direction to the location and the direction of the trajectory, divided by the number of angles measured. When the mean angle value is lower than 90°, the animal tends to orient itself towards the location; when the value is larger than 90°, it tends to avoid the location [34]. The median and quartiles of each distribution of 40 values were established.
Audacity

As in previous works [31-33], a cylindrical tower (height = 4 cm; diameter = 1.5 cm) set on a squared platform (9 cm²), both made of white Steinbach® paper, were deposited in the ants’ tray, and those present on this apparatus were counted 10 times over 10 minutes (Figure 2B). The counts obtained for the two colonies were added, and the mean and the extremes of these added counts were established. For statistical analysis, we used the counts obtained during two successive minutes (= 5 different counts).

Tactile perception

On a rough substrate, ants walk slowly, sinuously, with difficulties (Figure 2C). If they weakly perceive the uncomfortable character of such a substrate, they walk there more quickly and less sinuously. Therefore, for evaluating the ants’ tactile perception, we assessed their linear and angular speeds on a rough substrate (in the way explained above in ‘Linear and angular speeds’). As in previous works [31-33], the following apparatus was constructed for each colony. A folded piece (3 cm x 2 + 7 + 2 = 11 cm) of emery paper n° 280 was tied to the bottom and the borders of a tray (15 cm x 7 cm x 4.5 cm), which was so divided in a first 3 cm long zone, a second 3 cm long zone containing the emery paper; and a last 9 cm long zone. For making an experiment, 12 ants of each colony were deposited in the first zone of their apparatus, 24 trajectories of ants walking on the emery paper were recorded, and these ants’ linear and angular speeds were assessed. The median and quartiles of the obtained distributions of values were then established.

Brood caring behavior

A few larvae of each colony were removed from the nest and deposited in front of the entrance. Five of these larvae, and the ants’ behavior towards them, were observed (Figure 2D). The larvae, among the 5 + 5 = 10 observed, not re-entered in the nest after 5 seconds, 2, 4, 6, 8, and 10 minutes were counted.

Aggressiveness against nestmates and against aliens

Ants’ aggressiveness was assessed, as previously [31-33], in the course of 10 dyadic encounters between an ant (of colony A or B) and either a nestmate or an alien ant (Figure 2E and 2F). Each encounter occurred in a cylindrical cup (diameter = 2.5 cm, height = 1.8 cm) and lasted 5 minutes. During each of them, were counted the number of times the observed ant did nothing (level 0 of aggressiveness), contacted the opponent with its antennae (level 1), opened its mandibles (level 2), gripped the other ant (level 3), and tried to sting or stung the opponent (level 4). The numbers obtained during the 10 encounters were added. The ants’ aggressiveness was also evaluated by the variable “a” = n° of aggressiveness levels 2 + 3 + 4 / n° of levels 0 + 1.

Cognition

This protocol was set up while studying the effects of nicotine [35]. The following apparatus was built for each colony. Two pieces of duly folded white paper (Steinbach®, 12 cm x 4.5 cm) were inserted in a tray (15 cm x 7 cm x 4.5 cm) so divided into a first small zone, a zone with twists and turns, and a large zone containing a piece of wet cotton (Figure 2G). For performing an experiment, 15 ants of each colony were deposited in the first small zone of their apparatus, and the ants present in that zone and in the large one beyond the twists and turns were counted after 30 seconds, 2, 4, 6, 8, 10 and 12 minutes. The numbers obtained for the two colonies were added.

Escaping ability

For each colony, six ants were set under a reversed polyacetate glass (h = 8 cm, bottom diameter = 7 cm, ceiling diameter = 5 cm) deposited in their tray [31-33]. A notch made in the bottom rim of the glass (3 mm height, 2 mm broad) allowed the ants escaping (Figure 2H). The ants still enclosed and those escaped were counted after 30 seconds, 2, 4, 6, 8, 10 and 12 minutes, and the numbers obtained for the two colonies were added. The escape could also be evaluated by the proportion of ants escaped after 12 minutes.

Citation: Marie-Claire Cammaerts and Roger Cammaerts. “Adverse Effects of Etoricoxib, a Selectively Inhibiting Cyclo-Oxygenase-2 Anti-Inflammatory Drug, Assessed on Ants Used as a Model”. *EC Pharmacology and Toxicology* 7.5 (2019): 373-392.
Adverse Effects of Etoricoxib, a Selectively Inhibiting Cyclo-Oxygenase-2 Anti-Inflammatory Drug, Assessed on Ants Used as a Model

Conditioning and memory

The control experiment had been made four months before on a similar colony (see the subsection ‘Collection and maintenance of ants’). The test experiment was made using exactly the same protocol as that used for the control and the study of diclofenac and meloxicam [16,17], except that a yellow hollow cube was used for the present experiment test and a green one for the control. Each hollow cube was set above the entrance of an ants’ sugar water tube. The ants went so through visual conditioning. Their conditioning score was evaluated over time by making tests as follows. For making a test, 10 ants of each colony were individually deposited in a Y-apparatus provided with a new yellow hollow cube in one of its branch. The Y-apparatus was made of strong white paper and was located in a separate tray (30 cm x 15 cm x 4 cm). The yellow cube was randomly located in the right or the left branch of it. We noted into which branch each ant moved. Moving into the branch containing the cube was considered as giving the correct response (Figure 2I). Each test furnished thus the response of 20 ants, what allowed calculating the proportion of correct responses, i.e. the ants’ conditioning score.

Adaptation to potential adverse effects of etoricoxib

The ants’ linear and angular speeds in their foraging area, as well as on a rough substrate, were again assessed after the ants consumed etoricoxib for 7 and 8 days respectively. The results were compared to the control one and to those corresponding to 1 and 3 days respectively of etoricoxib consumption.

Habituation to potential beneficial effects of meloxicam

No beneficial effect having been found, this drug characteristic could not be examined.

Dependence on etoricoxib consumption

Dependence was examined after the ants consumed the drug since 10 days. As in previous works [31-33], for each colony, 15 ants were deposited in a tray (15 cm x 7 cm x 5 cm) containing two tubes (h = 2.5 cm, diam. = 0.5 cm), one filled with pure sugar water; the other filled with the sugar solution of etoricoxib used during the entire experimental work. The tube containing the drug was set on the right in one tray, and on the left in the other tray (Figure 2J). The ants coming onto each tube were counted 15 times over 15 minutes and the counts corresponding to each kind of liquid were separately added.

Decrease of the effect of etoricoxib on the ants’ sinuosity of movement, after its consumption was stopped

This decrease was examined after the ants consumed etoricoxib for 17 days, using a previously set up protocol [31-33]. The ants received a fresh solution of the drug 12 hours before the weaning time. After these 12 hours, the ants’ sinuosity was assessed. Then, at t = 0h, weaning started: the solution of etoricoxib was replaced by sugar water. Since this time, the ants’ sinuosity was assessed over time as it had been on ants under normal diet, on ants consuming etoricoxib for one day, and on ants consuming the drug for 7 days. The results are numerically given in table 5 and graphically presented in figure 4. The experiment ended when the ants’ sinuosity became similar to that presented under normal diet.

Statistical analysis

The numerical results concerning the ants’ linear speed, angular speed, orientation, tactile perception, aggressiveness against nest-mates and against aliens, adaptation, and decrease of the effects were ranked. Those obtained on ants under etoricoxib diet were compared to those obtained under normal diet using the non-parametric χ² test [36]. The ants’ food consumption, general activity, audacity, brood caring, cognition, escaping behavior; and conditioning capability, were statistically studied using the non-parametric test of Wilcoxon [36]. Ants’ dependence on etoricoxib consumption was analyzed using the non-parametric goodness of fit χ² test [36].

Citation: Marie-Claire Cammaerts and Roger Cammaerts. “Adverse Effects of Etoricoxib, a Selectively Inhibiting Cyclo-Oxygenase-2 Anti-Inflammatory Drug, Assessed on Ants Used as a Model”. EC Pharmacology and Toxicology 7.5 (2019): 373-392.
Adverse Effects of Etoricoxib, a Selectively Inhibiting Cyclo-Oxygenase-2 Anti-Inflammatory Drug, Assessed on Ants Used as a Model

The statistical analysis of the decrease of the effect of etoricoxib after weaning was made as follows. The distributions of the 20 sinuosity values obtained after given time periods were compared to that obtained at t = 0h and to the control one using the non-parametric $\chi^2$ test. Moreover, using Statistica V.10 software, the non-parametric Kruskal-Wallis two-tailed test (K-W test) for multiple comparisons was used to compare the values of sinuosity to either the values at the start of weaning (t = 0) or to the control values (the latter made at 20 values out of 40 in such a way that the median and quartiles were similar to those of the 40 values) [36]. A Bonferroni adjustment is incorporated. The mathematical function describing the regression of the median value of the sinuosity over time after weaning was established using Statistica® v.10 software, and the best choice between powers of polynomial regressions was established using the procedure described in Zar [37].

Results and Discussion

Food consumption, general activity

These physiological traits were affected by etoricoxib consumption (Table 1). Ants consuming this drug eat a little less meat than those not consuming it (mean values: 1.50 vs 1.86), but not statistically significantly (N = 6, T = +4, -17, P = 0.109). They drank somewhat less sugar water than ants living under normal diet (mean values: 1.86 vs 2.75), a difference statistically significant (N = 6, T = -21, P = 0.016). They also more often rested and were more inactive than ants living under normal diet (mean values: 8.16 vs 10.47), and this was also significant (N = 6, T = -21, P = 0.016). Etoricoxib impacted thus the ants’ food consumption (i.e. decreasing it), as well as their general activity (i.e. increasing in fact their resting time periods), what revealed some impact of the drug on the individuals’ physiology.

Linear and angular speeds

These traits were somewhat impacted by etoricoxib consumption (Table 2, lines 1, 2). Ants consuming this drug walked a little more quickly (a result only at the limit of significance: $\chi^2 = 7.06$, df = 3, 0.05 ~ P < 0.10), and more sinuously (a more significant result: $\chi^2 = 9.53$, df = 3, 0.02 ~ P < 0.05) than ants living under normal diet. This was obvious to the observer, as was obvious that their linear speed did not decrease and that their angular speed increased over their drug consumption. This last observation was confirmed after the ants had consumed etoricoxib during eight days (see below ‘Adaptation to adverse effects of etoricoxib’).

Orientation to an alarm signal

This trait was not all affected by etoricoxib consumption (Table 2, line 3; Figure 2A). Ants consuming the drug, just like those living under normal diet, well oriented themselves towards a tied nestmate (i.e. a source of attractive alarm pheromone). There was no statistical difference as for this ability between the ants maintained under one or the other kind of diet ($\chi^2 = 1.35$, df = 3, 0.70 < P < 0.80). Consequently, etoricoxib did not affect the ants’ olfactory perception as well as their ability in orienting themselves, what is in favor of the use of that drug.

Audacity

This trait was not at all affected by etoricoxib consumption (Table 2, line 3; Figure 2B). Indeed, ants consuming this drug reacted just like those living under normal diet when presented with an unknown risky apparatus. They came onto it, though somewhat hesitating, and did not stay there during a long time. There was not statistical difference between the ants maintained under one and the other kinds of diet as for the numbers of individuals seen over time on the risky apparatus (N = 4, T = +4.5, -5.5, P = 0.500). This lack of effect is in favor of the use of etoricoxib.

Tactile perception

Unexpectedly, this trait was accentuated by etoricoxib consumption (Table 2, the two last lines; Figure 2C). It was obvious to the observer that ants consuming this drug had some more difficulty in walking on the presented rough substrate than ants living under normal

Citation: Marie-Claire Cammaerts and Roger Cammaerts. “Adverse Effects of Etoricoxib, a Selectively Inhibiting Cyclo-Oxygenase-2 Anti-Inflammatory Drug, Assessed on Ants Used as a Model”. EC Pharmacology and Toxicology 7.5 (2019): 373-392.
diet. The numerical results reflected this observation. On such a substrate, the ants consuming etoricoxib moved at a lower linear speed and with a higher sinuosity than those not consuming this drug, and this was statistically significant (linear speed: $\chi^2 = 23.19$, df = 2, $P < 0.001$; angular speed: $\chi^2 = 14.60$, df = 2, $P < 0.001$). Etoricoxib increased thus, apparently, the ants’ tactile perception and/or sensitivity. This impact might exist for humans and should thus be examined in patients treated with etoricoxib. The ants’ locomotion on a rough substrate was again assessed after the ants consumed etoricoxib during eight days in order to examine if they could adapt themselves to this impact of the drug on their physiology. Even if this impact is far better than the inverse impact (i.e. a decrease of the individuals’ perception and/or sensitivity), it must not be too strong and optimally not long lasting for accepting the drug as an entirely safe one.

<table>
<thead>
<tr>
<th>Traits</th>
<th>Normal diet</th>
<th>Diet with etoricoxib</th>
</tr>
</thead>
<tbody>
<tr>
<td>Linear speed in mm/s</td>
<td>11.6 (10.5 - 12.5)</td>
<td>12.8 (11.7 - 14.4)</td>
</tr>
<tr>
<td>Angular speed in ang.deg/cm</td>
<td>121 (108 - 146)</td>
<td>150 (132 - 161)</td>
</tr>
<tr>
<td>Orientation in ang. deg.</td>
<td>32.4 (26.9 - 44.3)</td>
<td>34.0 (28.1 - 48.6)</td>
</tr>
<tr>
<td>Audacity (n°)</td>
<td>2.80 [1 - 4]</td>
<td>2.90 [2 - 4]</td>
</tr>
<tr>
<td>Tactile perception assessed on a rough substrate:</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Linear speed in mm/s</td>
<td>5.4 (4.6 - 6.5)</td>
<td>3.4 (2.9 - 4.0)</td>
</tr>
<tr>
<td>Angular speed in ang.deg/cm</td>
<td>254 (216 - 287)</td>
<td>332 (295 - 368)</td>
</tr>
</tbody>
</table>

Table 2: Impact of etoricoxib on five ethological and physiological traits. The drug increased the ants’ linear and angular speeds and their tactile perception, and did not impact their orientation and audacity. ang.deg.: Angular Degrees; ang.deg./cm: Angular Degrees Per cm; mm/s: Millimeters Per Second; n°: Numbers.

**Brood caring**

This ethological trait was not at all affected, and even somewhat improved by etoricoxib consumption (Table 3, line 1; Figure 2D). Ants consuming this drug as well as those living under normal diet rapidly tried to re-enter the larvae experimentally removed from the nest, but the former ants did so somewhat more promptly, a difference at the limit of significance (N = 4, T = -10, $P = 0.063$). Thus, etoricoxib did not affect the ants’ social tasks and relationships, a conclusion further checked in two following experiments (see below ‘Aggressiveness against nestmates’ and ‘Aggressiveness against aliens’).

**Aggressiveness against nestmates**

This ethological trait was not at all affected by etoricoxib consumption (Table 3, line 2 upper part; Figure 2E). Ants consuming this drug never aggressed the encountered nestmate, behaving just like ants living under normal diet ($\chi^2 = 3.82$, df = 2, 0.10 < $P < 0.20$). The ants’ social relationships were thus unchanged under etoricoxib diet, what was in agreement with the result concerning the brood caring behavior.

**Aggressiveness against aliens**

This trait was accentuated by etoricoxib consumption (Table 3, line 2, lower part, Figure 2F). Ants consuming this drug as ants not consuming it aggressed an alien, but the former less often opened their mandibles and more rapidly attacked, gripped and stung the opponent. The difference of behavior between ants living under one and the other kinds of diet was significant ($\chi^2 = 10.19$, df = 3, 0.01 < $P < 0.02$). Based on the observation of reacting ants, it could be presumed that ants consuming etoricoxib sooner (i.e. at a larger distance) perceived the alien character of the opponent and therefore more quickly attacked it. This presumed higher perception of ants consuming etoricoxib was in agreement with the observation of the more acute tactile perception of these ants (see above ‘Tactile perception’). The present experiment moreover showed that consuming a drug may affect (i.e. increase for instance) the consumer’s aggressiveness in some circumstances.
**Cognition**

Etoricoxib affected this physiological trait (Table 3, line 3; Figure 2G). Ants consuming this drug were less able to cross the difficult twist and turns path than ants not consuming it and this result was significant. For the numbers of ants still in front of the difficult path over time, the statistical result was $N = 7, T = +28, P = 0.008$; for the numbers of ants present over time beyond the difficult path, the result was $N = 5, T = -15, P = 0.031$. The ants’ cognitive abilities and/or brain functioning might thus be affected by etoricoxib, a presumption checked in a following experiment (see below ‘Conditioning ability, short and middle term memory’).

**Figure 2:** Some views of the experiments. 1: Ants under normal diet, 2: Ants under etoricoxib diet. A: Ants having reached a tied nestmate and having well succeeded under etoricoxib diet. B: Ants having come onto an unknown and risky apparatus. C: Ants walking with difficulties on a rough substrate. D: Ants taking care of a larva. E: Two nestmates presenting no aggressiveness. F: An ant stinging an alien one. G: Five ants under normal diet and two under etoricoxib diet (red circles) having succeeded in crossing a twists and turns path. H: An ant under normal diet escaping and two ants under etoricoxib diet not escaping from an enclosure. I: An ant trained to a green (normal diet) or a yellow cube (etoricoxib diet) giving the correct response when tested in a Y-apparatus provided with that cue in one of its branch.

**Escaping ability**

Etoricoxib impacted this ethological trait (Table 3, line 4). Under normal diet, enclosed ants walked all around in the enclosure, then essentially along its bottom rim, and so found the notch allowing escaping. Most of them went then out of the enclosure (i.e. ten among twelve ($= 0.83$) in 12 minutes). Ants consuming etoricoxib walked all round then along the rim, just like ants under normal diet, but, unexpectedly, seldom then going out of the enclosure. Only five among twelve ants went out over the 12 minutes of the test ($= 0.41$). This result was significant: $N = 6, T = +21$ (ants enclosed), $-21$ (ants escaped), $P = 0.016$. This might be due to some impact of the drug on the ants’ cognitive abilities (see the here above subsection 'Cognition'). The present experiment during which the ants’ escaping behavior was assessed was initially set up for evaluating the ants’ state of stress: the more the ants stressed, the lower was their ability in escaping.
Consequently, the result of the present experiment may indicate that etoricoxib somewhat increased the state of stress of the consumers (here the ants). On the other hand, several ants moving out of the enclosure, on the foraging area, were seen coming to the enclosure, perceiving their enclosed nestmates and trying to help them escaping. This observation was in agreement with the unchanged, excellent social relationships of ants consuming etoricoxib (see above ‘Brood caring’, and ‘Aggressiveness against nestmates’).

**Conditioning ability, short and middle term memory**

These traits were impacted by etoricoxib consumption (Table 3, last line; Figure 1). Under normal diet, ants easily acquired visual conditioning, reaching a score of 90% after 72 hours of training. While consuming etoricoxib, the ants poorly acquired such a conditioning, reaching a score of only 75% after 72 hours of training. This difference between ants living under one and the other kinds of diet was significant: \( N = 5, T = -15, P = 0.031 \). Etoricoxib impacted thus the animals’ ability of associating a cue and a reward (= the ability of becoming conditioned to a cue). Moreover, since the experiment lasted 72 hours, a rather long time in the course of which the ants became only poorly conditioned, it could be presumed that their short-term memory was affected by the drug. After 72 hours of training, the cues were removed from the ants’ trays and a test was performed 4 hours later. The ants then presented a score of 70%. Normally, a delay of four hours has no consequence on the conditioning score. For instance, in the course of a similar experiment on ants under meloxicam diet, four hours after the cue removal, the conditioned ants presented the same 90% score they presented before the cue removal [17]. We could thus also presume that etoricoxib decreased (or otherwise impacted) not only the short-term but also the middle-term memory, a potential effect which is not in favor of the use of this anti-inflammatory drug. However, since the ants went on going to their food sites, their nest entrance, and their cemeteries sites, their long-term memory may have been not affected by etoricoxib consumption.

<table>
<thead>
<tr>
<th>Traits</th>
<th>Normal diet</th>
<th>Diet with etoricoxib</th>
</tr>
</thead>
<tbody>
<tr>
<td>Brood caring: ( n^o ) of larvae not re-entered over 10 minutes</td>
<td>t: 30&quot;' 2' 4' 6' 8' 10'</td>
<td></td>
</tr>
<tr>
<td></td>
<td>10 7 4 3 1 0</td>
<td>8 6 4 2 0 0</td>
</tr>
<tr>
<td>Aggressiveness: levels and variable ‘a’</td>
<td>Levels: 0 1 2 3 4 'a'</td>
<td></td>
</tr>
<tr>
<td>Against nestmates (n°)</td>
<td>84 71 17 0 0 0.11</td>
<td>50 68 10 0 0 0.11</td>
</tr>
<tr>
<td>Against aliens (n°)</td>
<td>5 24 58 46 22 4.34</td>
<td>9 27 44 43 45 3.67</td>
</tr>
<tr>
<td>Cognition: ( n^o ) of ants in front (f) and beyond (b) a difficult path over 12 minutes</td>
<td>t: 30&quot;' 2' 4' 6' 8' 10' 12'</td>
<td></td>
</tr>
<tr>
<td></td>
<td>f: 28 22 17 14 12 12 11</td>
<td>29 25 20 17 16 16 14</td>
</tr>
<tr>
<td></td>
<td>b: 0 0 1 2 4 4 6</td>
<td>0 0 0 1 2 2 3</td>
</tr>
<tr>
<td>Escaping behavior: ( n^o ) of ants in and out of the enclosure over 12 minutes</td>
<td>t: 30&quot;' 2' 4' 6' 8' 10' 12'</td>
<td></td>
</tr>
<tr>
<td></td>
<td>in: 12 10 8 7 5 4 2</td>
<td>12 12 11 10 9 9 7</td>
</tr>
<tr>
<td></td>
<td>out: 0 2 4 5 7 8 10</td>
<td>0 0 1 1 3 3 5</td>
</tr>
<tr>
<td>Conditioning ability: score (in %) over time (in hours)</td>
<td>t: 7 24 31 48 55 72</td>
<td>7 24 31 48 55 72</td>
</tr>
<tr>
<td></td>
<td>score: 60 70 75 80 85 90</td>
<td>55 65 75 75 80 75</td>
</tr>
</tbody>
</table>

**Table 3:** Impact of etoricoxib on six physiological and ethological traits. The drug did not impact the ants’ social relationships (i.e. brood caring, aggressiveness) but affected their cognition, escaping behavior, and conditioning (and memory: see the text). \( n^o = \) Number, \% = Proportion, Levels: 0: Doing nothing, 1: Contacting the opponent with the antennae, 2: Opening the mandibles, 3: Gripping the opponent, 4: Stinging the opponent, \( t = \) Time.

**Citation:** Marie-Claire Cammaerts and Roger Cammaerts. “Adverse Effects of Etoricoxib, a Selectively Inhibiting Cyclo-Oxygenase-2 Anti-Inflammatory Drug, Assessed on Ants Used as a Model”. EC Pharmacology and Toxicology 7.5 (2019): 373-392.
Adverse Effects of Etoricoxib, a Selectively Inhibiting Cyclo-Oxygenase-2 Anti-Inflammatory Drug, Assessed on Ants Used as a Model

Adaptation to the impact of etoricoxib on locomotion

Ants consuming etoricoxib did not adapt themselves to the impact of this drug on their locomotion (Table 4, adaptation, upper part; Figure 3A). After having consumed etoricoxib for seven days, the ants still walked more quickly than while living under normal diet, and contrary to what occurred after one day of consumption, the increase was significant ($\chi^2 = 28.76$, df = 2, $P < 0.001$). Thus, the ants significantly walked more quickly after seven days than after one day of etoricoxib consumption ($\chi^2 = 10.76$, df = 2, $0.001 < P < 0.01$). Also, after having consumed etoricoxib for seven days, the ants continued to walk more sinuously than usually ($\chi^2 = 17.25$, df = 2, $P < 0.001$), and even slightly but not significantly more sinuously than after one day of that drug consumption ($\chi^2 = 2.65$, df = 2, $0.20 < P < 0.30$). This lack of adaptation to the impact of etoricoxib on the locomotion is not in favor of the use of this drug.

<table>
<thead>
<tr>
<th>Adaptation</th>
<th>Normal diet</th>
<th>Etoricoxib diet since 1 day</th>
<th>Etoricoxib diet since 7 days</th>
</tr>
</thead>
<tbody>
<tr>
<td>Linear speed in mm/sec</td>
<td>11.6 (10.5 - 12.5)</td>
<td>12.8 (11.7 - 14.4)</td>
<td>14.2 (13.2 - 15.1)</td>
</tr>
<tr>
<td>Angular speed in ang.deg./cm</td>
<td>121 (108 - 146)</td>
<td>150 (132 - 161)</td>
<td>158 (139 - 178)</td>
</tr>
</tbody>
</table>

<table>
<thead>
<tr>
<th>Adaptation to the impact on tactile perception assessed on a rough substrate:</th>
<th>Normal diet</th>
<th>Etoricoxib diet since 3 days</th>
<th>Etoricoxib diet since 8 days</th>
</tr>
</thead>
<tbody>
<tr>
<td>Linear speed in mm/sec</td>
<td>5.4 (4.6 - 6.5)</td>
<td>3.4 (2.9 - 4.0)</td>
<td>4.2 (3.5 - 4.8)</td>
</tr>
<tr>
<td>Angular speed in ang.deg./cm</td>
<td>254 (216 - 287)</td>
<td>332 (295 - 368)</td>
<td>334 (318 - 374)</td>
</tr>
</tbody>
</table>

Table 4: Adaptation to the impact of etoricoxib on locomotion and tactile perception, and dependence on this drug consumption. Adaptation: no adaptation occurred as for the impact of the drug on the ants’ locomotion and tactile perception. Dependence: ants presented no dependence on etoricoxib consumption. mm/sec: Millimeters Per Second; ang.deg./cm: Angular Degrees Per Centimeter.

Adaptation to the impact of etoricoxib on tactile perception

Ants did not adapt themselves to the impact of etoricoxib on their tactile perception (Table 3, adaptation, lower part; Figure 3B), which stayed still somewhat increased. This lack of adaptation was statistically significant. The linear speed on a rough substrate of ants having consumed etoricoxib for 8 days was still smaller than that of ants having not consumed this drug ($\chi^2 = 10.05$, df = 2, $0.001 < P < 0.01$), though slightly higher than that after 3 days of consumption ($\chi^2 = 5.68$, df = 1, $P = 0.02$). The sinuosity on a rough substrate of ants having consumed etoricoxib for 8 days was still higher than that of ants having not consumed this drug ($\chi^2 = 24.46$, df = 2, $P < 0.001$), and identical to that after 3 days of consumption ($\chi^2 = 0$, df = 1, NS). No adaptation occurred thus as for the impact of etoricoxib on the tactile perception and/or sensitivity, what might be an impediment to the use of this drug.

Dependence of etoricoxib consumption

Ants developed no dependence on etoricoxib consumption (Table 4, Dependence; Figure 3C). During the experiment, 36 ants of colony A (= 52.2%) went onto the tube containing the drug and 33 ones (= 47.8%) on the tube not containing it. At the same time, 31 ants of colony B (= 57.4%) went onto the tube free of the drug and 23 ones (= 42.6%) on the tube containing it. In total, 59 ants (= 48%) chose the liquid containing the drug and 64 ants (= 52%) chose the liquid free of it, a result not statistically different from that expected if ants randomly chose each kind of liquid (61.5, 61.5) ($\chi^2 = 0.016$, df = 1, $P = 0.90$). This experiment was repeated (the results being here not...
reported) with exactly the same absence of tendency to prefer the liquid containing the drug. Consequently, etoricoxib did not lead to
dependence, what is favorable to its use. During this experiment (and this is obvious on the photos presented in figure 3C), it could be
checked that the ants (living under etoricoxib diet) were not inclined in drinking lots of sugar water, an observation already made at the
start of the present study (see above ‘Food consumption, general activity’).

**Figure 3:** A: ants’ trajectories, 1 under normal diet, 2 under etoricoxib diet since 1 day,
3 under etoricoxib diet since 7 days: the ants did not adapt to the impact of the drug on their sinuosity. B: ants’ trajectories on
a rough substrate, 1 under normal diet, 2 under etoricoxib diet since 3 days, 3 under etoricoxib diet since 8 days: the ants did
not adapt to the impact of the drug on their tactile perception. C: ants’ dependence on etoricoxib consumption. The drug, present in the
tube with a red spot, did not attract the ants which thus became not dependent on it. Numerical results are given in Table 4.

**Figure 4:** Decrease of the effects of etoricoxib after weaning. Numerical results are given in table 5; Statistics are given in the text and in
table 5. Etoricoxib slowly and linearly lost its effect after weaning. It kept its initial effect until 12h - 13h after weaning and presented an
effect statistically not different from the control since about 16h after weaning. The ants’ sinuosity became identical to that under normal
diet 25h after weaning. ang.deg./cm: Angular Degrees/Centimeter, h: Hours.
Decrease of the effects of etoricoxib after weaning was stopped

The effect of etoricoxib slowly vanished after weaning (numerical results in Table 5, graphical presentation in Figure 4). The comparison of the values of ants’ sinuosity obtained over time after weaning with those obtained just before the start of weaning ($t = 0$) allowed stating that this drug kept its effect until about 12 hours after weaning. Indeed, the statistical results were as follows: 4 hours after weaning: $\chi^2 = 0.45$, df = 2, $P = 0.80$; 7 hours after weaning: $\chi^2 = 3.84$, df = 2, $0.10 < P < 0.20$; 10 hours after weaning: $\chi^2 = 0.18$, df = 2, $0.90 < P < 0.95$. After that, etoricoxib continued to lose its effect which became slightly lower than its initial one 13 hours after weaning ($\chi^2 = 7.38$, df = 2, $0.02 < P < 0.05$), then more and more lower 16 hours ($\chi^2 = 14.70$, df = 2, $P < 0.001$) and 19 hours ($\chi^2 = 20.08$, df = 2, $P < 0.001$) after weaning.

To evaluate if etoricoxib still had some effect beyond 13 hours after weaning, the values obtained over time were compared to the control ones (i.e. those obtained on ants under normal diet). Of course, the values obtained 4, 7, 10 and 13 hours after weaning highly statistically differed from the control ones ($P < 0.001$). Sixteen hours after weaning, the values differed from the control ones, being at the limit of significance ($\chi^2 = 5.97$, df = 2, $P ~ 0.05$), but 19 hours after weaning, they became different from the control ones ($\chi^2 = 8.48$, df = 2, $0.01 < P < 0.02$). Thereafter, 22 hours after weaning, the obtained values did not statistically differ from the control ones ($\chi^2 = 4.83$, df = 2, $0.05 < P < 0.10$), though being not yet equal to them. Beyond 22 hours after weaning, the values of ants’ sinuosity went on decreasing, and became similar to the control ones 25 hours ($\chi^2 = 0.16$, df = 2, $0.90 < P < 0.95$) and 31 hours ($\chi^2 = 2.46$, df = 2, $0.20 < P < 0.30$) after weaning. Etoricoxib became thus inefficient at about 21-23 hours after weaning.

Kruskal-Wallis tests on the values reflecting the decrease of the effect of etoricoxib after weaning leaded to similar conclusions. Compared to the values obtained at $t = 0$, those obtained after weaning became at the limit of significance 13 hours after weaning, and highly different beyond this time. Compared to the control values, those obtained after weaning stayed statistically different until 13 hours after weaning, then from 16 hours onwards they became not different. Compared to normal diet, etoricoxib appeared thus inefficient beyond 16 hours after weaning.

Table 5: Decrease of the effects of etoricoxib after weaning. More details are given in the text. The median values of sinuosity over time are graphically presented in Figure 4. Until 12-13 hours after weaning, etoricoxib kept its initial efficiency. From 16 hours after weaning, its effect became progressively undistinguishable from the control. Etoricoxib lost its effect slowly, linearly in a total of about 25 hours. ang.deg/cm: Angular Degrees/cm, $\chi^2$: Chi-Square; KW: Kruskal-Wallis tests.

<table>
<thead>
<tr>
<th>Time (hours)</th>
<th>Sinuosity ang.deg./cm</th>
<th>$\chi^2$ tests</th>
<th>KW test</th>
<th>vs t = 0</th>
<th>vs control</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>vs t = 0</td>
<td>vs control</td>
<td></td>
<td></td>
</tr>
<tr>
<td>0 = weaning</td>
<td>175 (164 – 186)</td>
<td>-</td>
<td>P &lt; 0.001</td>
<td>P &lt; 0.002</td>
<td></td>
</tr>
<tr>
<td>4</td>
<td>165 (156 – 185)</td>
<td>$P = 0.80$</td>
<td>$P &lt; 0.001$</td>
<td>$P = 1$</td>
<td>$P &lt; 0.002$</td>
</tr>
<tr>
<td>7</td>
<td>161 (148 – 182)</td>
<td>$0.10 &lt; P &lt; 0.20$</td>
<td>$P &lt; 0.001$</td>
<td>$P = 1$</td>
<td>$P &lt; 0.002$</td>
</tr>
<tr>
<td>10</td>
<td>164 (140 – 170)</td>
<td>$0.90 &lt; P &lt; 0.95$</td>
<td>$P &lt; 0.001$</td>
<td>$P = 0.430$</td>
<td>$P &lt; 0.002$</td>
</tr>
<tr>
<td>13</td>
<td>150 (135 – 165)</td>
<td>$0.02 &lt; P &lt; 0.05$</td>
<td>$P &lt; 0.001$</td>
<td>$P = 0.054$</td>
<td>$P = 0.012$</td>
</tr>
<tr>
<td>16</td>
<td>145 (128 – 152)</td>
<td>$P &lt; 0.001$</td>
<td>$P ~ 0.05$</td>
<td>$P = 0.002$</td>
<td>$P = 0.258$</td>
</tr>
<tr>
<td>19</td>
<td>135 (123 – 148)</td>
<td>$P &lt; 0.001$</td>
<td>$0.01 &lt; P &lt; 0.02$</td>
<td>$P &lt; 0.001$</td>
<td>$P = 1$</td>
</tr>
<tr>
<td>22</td>
<td>137 (121 – 129)</td>
<td>$P &lt; 0.001$</td>
<td>$0.05 &lt; P &lt; 0.10$</td>
<td>$P &lt; 0.001$</td>
<td>$P = 1$</td>
</tr>
<tr>
<td>25</td>
<td>121 (108 – 148)</td>
<td>$P &lt; 0.001$</td>
<td>$0.90 &lt; P &lt; 0.95$</td>
<td>$P &lt; 0.001$</td>
<td>$P = 1$</td>
</tr>
<tr>
<td>31</td>
<td>117 (93 –137)</td>
<td>$P &lt; 0.001$</td>
<td>$0.20 &lt; P &lt; 0.30$</td>
<td>$P &lt; 0.001$</td>
<td>$P = 1$</td>
</tr>
<tr>
<td>Control</td>
<td>121 (108 – 146)</td>
<td>$P &lt; 0.001$</td>
<td>-</td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

Citation: Marie-Claire Cammaerts and Roger Cammaerts. “Adverse Effects of Etoricoxib, a Selectively Inhibiting Cyclo-Oxygenase-2 Anti-Inflammatory Drug, Assessed on Ants Used as a Model”. *EC Pharmacology and Toxicology* 7.5 (2019): 373-392.
The decrease of the effect of etoricoxib was slow and linear, its function being:

\[ E_t = 175,786 - 1,955 t \]

with \( E_t \) = effect (median sinuosity) at time \( t \); \( t \) = time since weaning (h)

Such a slow linear decrease is in favor of the use of etoricoxib because it allows consuming only a small amount of the drug and avoiding dependence.

Discussion

Anti-inflammatory drugs are among the most consumed drugs and are a medical necessity. Traditional NSAIDs (e.g. diclofenac) present several adverse effects, the most frequent and painful one being stomach impairment. A new generation of anti-inflammatory drugs has thus been elaborated [10,11]. These ‘coxib’ drugs inhibit more specifically the COX2 enzyme [6,38]. Consequently, they are better tolerated by the stomach although efficient in treating rheumatologic and intestinal inflammatory reactions [13, 15]. However, they were suspected or appeared to present adverse effects, essentially vascular and maybe kidneys problems [7,11,14,39-41]. Due to their recent use by patients, to the small amount of experiments made with them and to the still incomplete understanding of the role of COX-2, the long-term use of high doses of these coxibs should be limited whenever as possible [42].

Having examined, on ants used as models, the effects of two traditional anti-inflammatory drugs, diclofenac [16] and meloxicam [17], we here examined in the same way the effects of a coxib of the second generation, etoricoxib. Here below, we compare the safety of diclofenac, meloxicam and etoricoxib on the basis of (1) our results on ants, (2) the instructions for used supplied with the drugs, (3) studies made on vertebrates and humans. This comparison is summarized in table 6.

<table>
<thead>
<tr>
<th>Source</th>
<th>Diclofenac, previous work [16]</th>
<th>Meloxicam, previous work [17]</th>
<th>Etoricoxib, present work</th>
</tr>
</thead>
<tbody>
<tr>
<td>Our works on ants</td>
<td>Meat consumption decreased</td>
<td>Meat consumption decreased</td>
<td>Meat consumption decreased</td>
</tr>
<tr>
<td></td>
<td>Sugar water intake decreased</td>
<td>Sugar water intake increased</td>
<td>Sugar water intake decreased</td>
</tr>
<tr>
<td></td>
<td>General activity decreased</td>
<td>General activity decreased</td>
<td>General activity decreased</td>
</tr>
<tr>
<td></td>
<td>Linear speed unchanged</td>
<td>Linear speed unchanged</td>
<td>Linear speed increased</td>
</tr>
<tr>
<td></td>
<td>Sinuosity increased</td>
<td>Sinuosity unchanged</td>
<td>Sinuosity increased</td>
</tr>
<tr>
<td></td>
<td>Orientation ability decreased</td>
<td>Orientation ability unchanged</td>
<td>Orientation ability unchanged</td>
</tr>
<tr>
<td></td>
<td>Audacity decreased</td>
<td>Audacity decreased</td>
<td>Audacity unchanged</td>
</tr>
<tr>
<td></td>
<td>Tactile perception unchanged</td>
<td>Tactile perception unchanged</td>
<td>Tactile perception increased</td>
</tr>
<tr>
<td></td>
<td>Brood caring unchanged</td>
<td>Brood caring unchanged</td>
<td>Brood caring unchanged</td>
</tr>
<tr>
<td></td>
<td>Social relationships unchanged</td>
<td>Social relationships improved</td>
<td>Social relationships unchanged</td>
</tr>
<tr>
<td></td>
<td>Cognition decreased</td>
<td>Cognition decreased</td>
<td>Cognition decreased</td>
</tr>
<tr>
<td></td>
<td>Escaping ability decreased</td>
<td>Escaping ability decreased</td>
<td>Escaping ability decreased</td>
</tr>
<tr>
<td></td>
<td>Conditioning decreased</td>
<td>Conditioning unchanged</td>
<td>Conditioning decreased</td>
</tr>
<tr>
<td></td>
<td>Memorization decreased</td>
<td>Memorization unchanged</td>
<td>Memorization decreased</td>
</tr>
<tr>
<td></td>
<td>No adaptation</td>
<td>No adaptation</td>
<td>No adaptation</td>
</tr>
<tr>
<td></td>
<td>Some dependence</td>
<td>No dependence</td>
<td>No dependence</td>
</tr>
<tr>
<td></td>
<td>Rapid decrease of effects after weaning</td>
<td>No decrease for 6 hours, then slow decrease of effects after weaning</td>
<td>Slow decrease of effects after weaning</td>
</tr>
</tbody>
</table>
We found that etoricoxib decreased the ants’ meat consumption, sugar water consumption, activity, cognition, escaping ability, conditioning, and memory, that it increased the ants’ linear and angular speeds, state of stress, and tactile perception. No adaptation occurred to these effects. The ants’ orientation, audacity, brood caring and relation with nestmates were not affected by etoricoxib. The ants sooner aggressed an alien, what points out a possible impact of a drug on the individual aggressiveness. Ants developed no dependence on this drug consumption. The effects of etoricoxib slowly and linearly decreased in about 21 - 23 hours after weaning. Tested on ants, the new anti-inflammatory drug presented thus some adverse effects. According to the examined social relationships and memorization traits, etoricoxib appeared not safer than meloxicam, and concerning orientation ability, audacity and dependence, it appeared safer than diclofenac.

As for the instructions for used supplied with the drugs, their list of adverse effects are rather similar. Nevertheless, meloxicam appears to present somewhat less numerous, less frequent and less important adverse effects, leading among others to few cardiovascular risks.

Concerning studies on vertebrates, those relative to etoricoxib are less numerous and less developed than those relative to diclofenac and meloxicam because the use of etoricoxib is more recent. Agrawal and co-workers [43] showed that, in healthy humans, etoricoxib is well tolerated across the whole pharmaceutical dose range and that its pharmacokinetics is linear with a half-life of 22 hours. In the present work on ants, we also observed a long duration of the effects of this drug. Agrawal, et al briefly mentioned that the COX2 enzyme is

Table 6: Comparison of the adverse effects of diclofenac, meloxicam and etoricoxib, three anti-inflammatory drugs, on the basis of our works on ants, the instructions joined to the drug package, and research made on vertebrates.

In the diclofenac and meloxicam vertebrate research section (columns 2 and 3, last part), the numbers into brackets refer to the papers listed in the bibliography of our preceding works [16,17] and not to the numbers of the present bibliography. Moreover, for these sections, the report of Lapeyre-Mestre., et al. [58] is also taken into account.
constitutively expressed in brain and kidneys, but did not examine these organs in patients treated with etoricoxib. In the present study on ants, we observed some slight effect of the drug on the brain. Reviewing the pharmacologic properties of etoricoxib, Takemoto., et al [44] found that etoricoxib is effective, has a lower gastrointestinal incidence than traditional NSAIDs such as diclofenac, induces the same risk of cardiovascular problems as diclofenac, and leads to no more renal adverse events than other coxibs, except rofecoxib. They conclude that etoricoxib has similar efficiency as traditional NSAIDs, but requires further examination as for cardiovascular adverse effects and that it still remains to determine if its benefits outweigh its risks. Nevertheless, etoricoxib has been considered as relatively safe as for its impact on hematological parameters, colon and kidneys by Behal., et al [45]. Be that it may, etoricoxib was proved to be more effective as an analgesic against dental pain than e.g. ibuprofen or paracetamol/codein [44], against postoperative periodontal pain [46] and against osteoarthritis pain [47]. We did not assess the effect of etoricoxib on pain in ants, but showed that this drug enhanced their tactile sensitivity, while diclofenac and meloxicam did not [16, 17].

Among other interesting effects of etoricoxib, it has been demonstrated that it prevented alveolar bone loss in periodontitis [48, 49] and has a potential protective effect in colon and lung cancer [50-53]. However, there are some rare cases of hypersensitivity to etoricoxib, leading to immediate urticaria, acute generalized pustulosis and toxic epidermal necrolysis [54-57].

Etoricoxib is thus an effective coxib, although not without adverse effects, some of them having probably been not yet published or even remaining to be discovered. The present work on ants shows that this drug affects at least tactile sensitivity, cognition and memorization.

**Conclusion**

Tested on ants used as a model, the anti-inflammatory drug etoricoxib presents some adverse physiological and behavioral effects, among them, an increase of tactile sensitivity and a decrease of cognition and memorization. Compared to other anti-inflammatory drugs, etoricoxib appears to be safer than diclofenac which impacts the ants’ orientation and audacity and induces some dependence. However, etoricoxib appears not to be safer than meloxicam which does not affect the ants’ tactile perception, conditioning ability and memorization. Our conclusion about the safety of etoricoxib examined on ant traits is that it is less safe than meloxicam and safer than diclofenac. Other kind of information on the safety of the drugs are provided by the instructions for human use joined to their package, as well as by the results of research made on vertebrate animals and humans. This information indicates that etoricoxib, although relatively safe for kidney and colon, may induce oedema and more serious skin problems than meloxicam and diclofenac, and that the latter induces more kidney and gastrointestinal problems. Therefore, according to our knowledge of the impacts of etoricoxib, meloxicam and diclofenac on ants and on vertebrates, meloxicam appears to be the safest of these drugs.

**Conflict of Interest**

We affirm having no conflict of interest as for the use of etoricoxib, or any other anti-inflammatory drugs. We are biologists and ethologists, working on the behavior and the physiology of ants and their societies. We receive no money for making our research.

**Bibliography**

Adverse Effects of Etoricoxib, a Selectively Inhibiting Cyclo-Oxygenase-2 Anti-Inflammatory Drug, Assessed on Ants Used as a Model


Citation: Marie-Claire Cammaerts and Roger Cammaerts. “Adverse Effects of Etoricoxib, a Selectively Inhibiting Cyclo-Oxygenase-2 Anti-Inflammatory Drug, Assessed on Ants Used as a Model”. EC Pharmacology and Toxicology 7.5 (2019): 373-392.
Adverse Effects of Etoricoxib, a Selectively Inhibiting Cyclo-Oxygenase-2 Anti-Inflammatory Drug, Assessed on Ants Used as a Model


Adverse Effects of Etoricoxib, a Selectively Inhibiting Cyclo-Oxygenase-2 Anti-Inflammatory Drug, Assessed on Ants Used as a Model


Citation: Marie-Claire Cammaerts and Roger Cammaerts. "Adverse Effects of Etoricoxib, a Selectively Inhibiting Cyclo-Oxygenase-2 Anti-Inflammatory Drug, Assessed on Ants Used as a Model". *EC Pharmacology and Toxicology* 7.5 (2019): 373-392.