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Anne-Laure Pélissier-Alicot · Jean-Michel Gaulier · Carine Dupuis · Marc Feuerstein · Georges Léonetti · Gérard Lachâtre · Pierre Marquet

Post-mortem redistribution of three beta-blockers in the rabbit

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Abstract To consider the role of the physico-chemical properties of drugs in their post-mortem redistribution, we designed the present study to investigate the influence of lipophilicity using an experimental rabbit model. Three beta-blockers (BB), atenolol, metoprolol and propranolol, with a similar dissociation constant (pK_a) and increasing partition coefficient (K_p) were administered intravenously to 18 rabbits. One hour after the last administration, the animals were killed by thiopental injection and placed in a supine position at room temperature. Autopsies were performed at 0, 2, 6, 12, 24 and 48 h post-mortem. Concentrations of the three BB were determined in fluids (right and left cardiac blood, peripheral blood, urine, bile, stomach content, vitreous humour) and tissues (cardiac muscle, lungs, liver, brain, diaphragm, iliopsoas muscle) using a previously published, validated liquid chromatography–electrospray–mass spectrometry method. Our results show that lipophilicity influences post-mortem redistribution of the molecules in a certain number of anatomical sites such as the stomach, lungs, cardiac muscle, cardiac blood or liver, but does not appear to intervene in other sites such as the brain or the vitreous humour.

Keywords Beta-blockers · Post-mortem redistribution · Lipophilicity · K_p · Rabbits

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A.-L. Pélissier-Alicot (✉) · G. Léonetti
Service de Médecine Légale, Faculté de Médecine,
27, boulevard Jean Moulin,
13385 Marseille, Cedex 5, France
e-mail: apelissier@ap-hm.fr
Tel.: +33-4-91324516
Fax: +33-4-92324512

J.-M. Gaulier · C. Dupuis · G. Lachâtre · P. Marquet
Laboratoire de Pharmacologie et Toxicologie, CHU Dupuytren,
2, avenue Martin Luther King,
87042 Limoges, Cedex, France

M. Feuerstein
Centre de Formation et de Recherches Experimentales
Médico-chirurgicales, Faculté de Médecine,
27, boulevard Jean Moulin,
13385 Marseille, Cedex 5, France

G. Lachâtre
Laboratoire de Toxicologie, Faculté de Pharmacie,
2 rue du Docteur Marcland,
87025 Limoges, Cedex, France

Introduction

Variations in drug concentrations in the post-mortem period lead to numerous difficulties in the interpretation of results [1]. The redistribution mechanisms of xenobiotics from reservoir organs (stomach, lungs, liver, cardiac muscle) have been extensively described [2–7]. Similarly, variations in concentrations related to cell lysis and putrefaction have been the object of several studies [8, 9]. However, the influence of the physico-chemical and pharmacokinetic parameters of xenobiotics is not well-known. Only the influence of the volume of distribution (V_d) of a molecule on its potential redistribution has been studied, and the results tend to show that molecules with a wide V_d are those which are most intensely affected by post-mortem redistribution because of their extensive tissue uptake [10]. These are generally weak, basic lipophilic drugs for which a phenomenon of intracellular ion trapping intervenes [11, 12]. This has been extensively demonstrated for tricyclic antidepressants, amphetamine and methamphetamine, and digoxin [13–15]. According to Hilberg et al. [16], all molecules with a V_d equal to or greater than 3 L/kg are liable to undergo post-mortem redistribution. However, this hypothesis cannot account for the phenomena of redistribution observed with all molecules. Indeed, several examples demonstrate that the post-mortem site- and time-dependent variations of drug concentrations are not systematically

associated with molecules with a high apparent V_d. Acetaminophen, which has a small apparent V_d of about 1 L/kg, presents site- and time-dependent variations in post-mortem blood levels, with higher concentrations in cardiac than in femoral blood at each sampling time, and with an increase in cardiac blood levels over time [17, 18]. The opposite situation is illustrated by mirtazapine, a relatively new antidepressant with basic and lipophilic properties and an apparent V_d of 4.8 L/kg, which did not exhibit any significant difference between cardiac and femoral post-mortem blood concentrations, though it showed an increase in liver concentrations [19–21]. It would thus seem that the redistribution of a molecule must be interpreted in accordance with a range of physico-chemical and pharmacokinetic parameters and not only as a function of its lipophilicity and/or its V_d. If general rules governing these phenomena could be defined, based on physico-chemical and pharmacokinetic parameters, this would facilitate the interpretation of post-mortem redistribution.

The aim of this work was to study the influence of the partition coefficient (K_p) on the post-mortem redistribution in rabbits of three beta-blockers (BB) of toxicological interest, namely, atenolol, metoprolol and propranolol, with similar dissociation constant (pK_a) and increasing K_p [22].

Materials and methods

The principles of laboratory animal care (NIH publication No. 85-23, revised 1985) were followed, and the study protocol was approved by the Ethics Committee on Animal Experimentation of the Medical Faculty, Mediterranean University of Marseille (request number 28/02).

Animals

The animal model chosen was the rabbit, essentially for two reasons. The first is that this animal has a gall bladder and that bile is a medium of interest in medico-legal toxicology, in particular for molecules with an enterohepatic cycle, which is the case for propranolol [23]. The second reason is that the vitreous humour of the rabbit has similar chemical characteristics to that of man [24].

Eighteen 12-week-old male New Zealand rabbits, with a mean weight of 2200 g, were purchased from ESD (Bourgen-Bresse, France). They were housed individually in steel cages under controlled environmental conditions, i.e. relative humidity of 55±5% and temperature of 22±2°C. Food and water were available ad libitum.

Beta-blockers

Atenolol, metoprolol and propranolol were obtained as pure substances from Sigma-Aldrich (Saint-Quentin Fallavier, France). Their respective pK_a and K_p values are reported in Table 1.

Table 1 Physico-chemical properties of the three beta-blockers [22]

	Atenolol	Metoprolol	Propranolol
K_p	0.01	0.18	5.39
pK_a	9.60	9.68	9.45
Protein binding (%)	<5	10	90
V_d (L/kg)	0.7	4	4

K_p partition coefficient, pK_a dissociation constant, V_d volume of distribution

Using dosing data found in the literature [25], increasing doses were tested intravenously on rabbits: a 5-mg/kg non-lethal dose was finally chosen. The mixture of the three BB at the final concentration of 5 mg/mL was prepared by dissolving 1.5 g of atenolol, metoprolol and propranolol obtained as pure substances, with the addition of 2.1 g of citric acid to make dissolution easier, in 300 mL of 0.9% NaCl. This mixture was filtered and conditioned into sealed vials under sterile conditions. The stability of this solution was verified at room temperature for 30 days.

Experiments

Three sets of experiments were performed (Table 2). In each experiment, six animals received the solution of the three BB at 5 mg/kg i.v. via the ear marginal vein three times in two days: the first day at 9.00 a.m. and 4.00 p.m., and the second day at 9.00 a.m. The animals were killed by an injection of thiopental (150 mg/kg i.v.) 1 h after the last administration to allow BB distribution. They were left in the supine position at 20°C until autopsy was performed at 0, 2, 6, 12, 24 and 48 post-mortem hours (one animal at a time). Right and left cardiac blood samples were drawn after clamping the inferior vena cava just above the diaphragm. Peripheral blood was drawn from the infrarenal inferior vena cava because the quantity of femoral venous blood is too small to allow drug determination, especially at late stages. Samples were also collected from the urine, bile, stomach content, vitreous humour, right and left

Table 2 Study design

Day	Time of injection	Time of killing	Sampling time (post-mortem interval)	
D0	0900 hours			
	1600 hours			
D1	0900 hours	1000 hours	1000 hours	(T0)
			1200 hours	(T2)
			1600 hours	(T6)
			2200 hours	(T12)
D2			1000 hours	(T24)
D3			1000 hours	(T48)

In each experiment (n=3), six animals received the solution of the three beta-blockers at a dose of 5 mg/kg i.v. Post-mortem intervals are expressed in hours

ventricles of the heart, right apex, right base, left apex and left base of the lungs, right and left lobes of the liver, brain, diaphragm and iliopsoas muscle. The blood samples were collected in tubes with 1% sodium fluoride. All samples were stored at −20°C until analysis.

Analytical methods

Concentrations of the three BB were determined using a previously published, fully validated liquid chromatography–electrospray–mass spectrometry (LC–ES–MS) method [26]. Briefly, after addition of clenbuterol, a beta-agonist used as internal standard, the samples were extracted from 2.0 mL of fluids or 200 mg of solid tissues by solid–liquid extraction using Extrelut columns. Chromatographic separation involved a Nucleosil C18, 5-μm (150×1 mm i.d.) column together with a gradient of acetonitrile in 2 mM, pH 3 ammonium formate. The compounds were ionised in the electrospray source of the mass spectrometer, fragmented by in-source collisions, and the fragment ions positively detected selected ion-monitoring mode, targeting one quantitation and two confirmation ions per compound. The limit of quantitation was 50 μg/L in blood, stomach content and urine, 10 μg/L in the vitreous humour and 50 ng/g in tissues. The technique was found to be linear between 50 and 5,000 ng/g in the heart and liver, between 50 and 5,000 μg/L in urine extracts, between 1,000 and 50,000 ng/g in the lung and kidney, and between 500 and 5,000 μg/L in the stomach content; a quadratic equation best fitted the measured and theoretical concentrations in blood between 50 and 5,000 μg/L, as well as in the brain between 50 and 40,000 ng/g.

Data analysis

Statistical processing of the data was performed using non-parametric tests (SPSS program V 11.0 for Windows). The Kruskal–Wallis test was applied to compare the concentrations of the three BB as a function of the post-mortem interval. The Wilcoxon rank test was used for the analysis of intra-individual differences in concentrations for each BB between right cardiac blood, left cardiac blood and peripheral blood, between right and left ventricles, right and left apexes, right and left bases of lungs and finally between diaphragm and iliopsoas muscle. The relationships between these values were investigated with the Spearman correlation test. For all tests, p values less than 0.05 were considered to be statistically significant.

Results

Measured concentrations are reported as mean±SD in Table 3. No data were reported at 48 h in peripheral blood as the sample volume obtained at this post-mortem interval was too small, and data for 12 h in bile were not available owing to a chromatographic run problem. Changes in relative concentrations (ratio of measured concentration/concentration at 0 h post-mortem) of the three BB in each matrix are shown in Fig. 1a and b in ESM.

At 0 h post-mortem, the atenolol concentration in peripheral blood was similar to the concentrations in the other fluid or tissue sampling sites. For metoprolol, only the concentrations in the lung apexes were higher than those in peripheral blood. On the other hand, for propranolol, peripheral blood concentration was markedly lower than the concentrations in cardiac blood and in the right and left cardiac muscle, right and left lung apexes and bases, brain, iliopsoas muscle and diaphragm.

In the various post-mortem intervals, atenolol, metoprolol and propranolol concentrations did not vary significantly in the samples of right cardiac, left cardiac and peripheral blood, with, however, a tendency to decrease, which was followed by an increase after 2 h for left cardiac and peripheral blood, and after 12 h for right cardiac blood. These results cannot be compared with later samplings because the three BB were not measured after 24 h in peripheral blood owing to the small blood volume obtained. Moreover, the Wilcoxon test showed no significant difference between the various blood compartments for each BB. In the right cardiac muscle, the Kruskal–Wallis test revealed a significant increase in propranolol (p=0.02), in particular at longer post-mortem intervals, whereas atenolol and metoprolol concentrations remained stable. In the left cardiac muscle, concentrations of propranolol and metoprolol increased significantly (p=0.04 and 0.03, respectively), with that of propranolol being more remarkable. However, no significant difference was observed between left and right cardiac muscle concentrations for the three BB with the Wilcoxon test. In the lung, concentrations of the three BB followed a similar course: atenolol concentrations remained stable, whereas metoprolol and propranolol concentrations decreased significantly (right lung apex: p=0.02 and 0.01, respectively; right lung base: p=0.02 and 0.01, respectively; left lung apex: p=0.03 and 0.02, respectively; left lung base: p=0.04 and 0.01, respectively). The concentrations of each BB were similar between both apexes and between both bases, but we observed significantly higher concentrations of metoprolol and propranolol in the apexes than in the right (p=0.01 and 0.02, respectively) and left bases (p=0.01 and 0.04, respectively), with these concentrations being correlated for both the right lung (r_s=0.935 and 0.892; p=0.04 and 0.02, respectively, for metoprolol and propranolol) and the left lung (r_s=0.843 and 0.708; p=0.03 and 0.04, respectively, for metoprolol and propranolol). In the stomach content, we observed no statistically significant difference between the various sampling times, although propranolol and, to a lesser extent, metoprolol concentrations peaked at 2 h, followed by a decrease until 12 h and then a new peak at 24 h. Atenolol followed the opposite course. Results were similar in urine. In both liver lobes, concentrations were similar and stable over time, although propranolol tended to increase slightly. In the vitreous humour, concentrations showed a non-significant increase with time for propranolol and a significant increase for metoprolol and atenolol (p=0.02 and

Table 3 Mean atenolol, metoprolol and propranolol concentrations in the different sampling sites (values expressed as mean±SD)

Samples (concentration units)	Atenolol						Metoprolol						Propranolol					
	T0	T2	T6	T12	T24	T48	T0	T2	T6	T12	T24	T48	T0	T2	T6	T12	T24	T48
Right cardiac blood (μg/L)	5790 (1830)	4181 (1043)	3521 (2368)	4030 (197)	4624 (578)	3441 (460)	1102 (212)	702 (169)	580 (376)	524 (75)	929 (130)	544 (401)	502 (101)	432 (64)	440 (419)	354 (151)	809 (300)	549 (48)
Left cardiac blood (μg/L)	6936 (2296)	3052 (309)	3861 (1074)	4214 (326)	4425 (1556)	5344 (5320)	1686 (316)	420 (130)	739 (194)	786 (20)	767 (317)	1147 (1106)	1220 (440)	304 (72)	595 (299)	545 (74)	472 (126)	825 (520)
Peripheral blood (μg/L)	5486 (2292)	1857 (984)	3364 (2904)	17706 (19552)	7632 (2441)		966 (392)	351 (120)	863 (623)	3416 (3770)	1390 (728)		376 (119)	190 (118)	480 (360)	731 (514)	817 (327)	
Right myocardium (ng/g)	5259 (3943)	5567 (429)	4562 (1767)	5166 (1927)	5398 (1452)	5961 (1079)	1495 (960)	1584 (435)	2184 (901)	1857 (822)	2060 (320)	3188 (231)	2864 (1731)	2812 (269)	3930 (676)	6161 (2537)	9687 (2120)	29840 (7559)
Left myocardium (ng/g)	2429 (740)	4194 (57)	4293 (749)	8932 (3611)	7312 (2087)	6585 (1745)	794 (167)	1330 (241)	2072 (568)	2609 (767)	3211 (721)	3656 (1502)	3355 (1867)	4284 (1739)	3822 (694)	4649 (818)	7986 (532)	24506 (15991)
Right apex lung (ng/g)	6103 (1766)	10487 (995)	2787 (1281)	4957 (939)	4639 (425)	4882 (870)	11733 (3659)	11032 (999)	3331 (1264)	5038 (842)	3056 (236)	3461 (880)	48353 (4757)	73890 (4247)	36773 (5112)	30153 (3863)	26480 (8083)	24090 (7730)
Left apex lung (ng/g)	6987 (1522)	6864 (2189)	3450 (285)	5698 (1523)	4523 (363)	5528 (1241)	14021 (3566)	9492 (1533)	3097 (964)	4695 (2382)	3268 (1727)	3406 (698)	56426 (2567)	59903 (12758)	32693 (3505)	28056 (14484)	19686 (3451)	22843 (5138)
Right base lung (ng/g)	6034 (1819)	8280 (2972)	3819 (1570)	4721 (581)	6051 (785)	5271 (1557)	9411 (4021)	10700 (3081)	2931 (1217)	4144 (972)	2053 (975)	2923 (699)	46543 (8090)	63320 (8770)	32920 (8658)	24703 (2014)	14749 (7706)	18653 (2362)
Left base lung (ng/g)	8059 (3211)	7199 (4305)	3321 (2761)	3699 (1024)	5348 (752)	4496 (784)	13710 (7950)	8266 (3595)	2273 (1046)	2440 (1433)	3019 (1211)	2566 (710)	45256 (4608)	37760 (8624)	28970 (5573)	17960 (2058)	22963 (3159)	15860 (3241)
Right liver lobe (ng/g)	6115 (1101)	5180 (1284)	4484 (1967)	4573 (748)	3939 (1159)	4804 (943)	1150 (513)	1126 (592)	1110 (427)	1060 (94)	1246 (509)	1487 (386)	287 (205)	286 (87)	476 (119)	497 (161)	651 (117)	541 (413)
Left liver lobe (ng/g)	5299 (1736)	3845 (1881)	5996 (1395)	4776 (1090)	4675 (1062)	3800 (505)	1048 (56)	722 (370)	1177 (212)	1077 (275)	1159 (340)	1853 (470)	295 (201)	280 (123)	575 (286)	341 (268)	518 (235)	462 (413)
Gastric fluid (μg/L)	1369 (880)	1449 (901)	2375 (2164)	3316 (107)	2332 (768)	3843 (825)	7682 (1719)	14993 (3180)	10709 (5158)	10189 (1872)	14250 (4009)	10939 (2752)	1863 (502)	7286 (1902)	6636 (4350)	4265 (1541)	4832 (446)	2768 (246)
Bile (μg/L)	34240 (24338)	25870 (17810)	24140 (2545)		9064 (1033)	7263 (6317)	3294 (1408)	2854 (1117)	3042 (1787)		1690 (1712)	1305 (340)	1270 (590)	503 (103)	512 (337)		1074 (679)	310 (111)
Urine (μg/L)	50520 (18794)	173900 (9050)	212693 (142514)	266100 (71038)	214500 (67950)	93180 (21665)	136 (15)	204 (88)	12890 (8006)	5168 (85)	3197 (2283)	3579 (2559)	7 (1)	15 (11)	1657 (2497)	88 (77)	815 (1210)	228 (153)
Brain (ng/g)	165 (48)	163 (46)	218 (156)	277 (115)	730 (673)	228 (42)	1382 (1309)	1938 (555)	1913 (978)	1910 (226)	2219 (249)	2310 (373)	11863 (5059)	11383 (1540)	7958 (1284)	8270 (2729)	8129 (2681)	12976 (2121)
Vitreous humour (μg/L)	65 (21)	124 (77)	154 (101)	231 (29)	776 (304)	991 (280)	271 (88)	432 (394)	391 (171)	555 (367)	1822 (511)	2760 (453)	55 (30)	341 (680)	91 (37)	168 (55)	545 (386)	615 (228)
Iliopsoas muscle (ng/g)	759 (365)	781 (119)	657 (217)	810 (10)	992 (145)	913 (235)	462 (241)	444 (79)	351 (178)	322 (58)	348 (25)	380 (174)	602 (362)	580 (137)	499 (377)	336 (148)	347 (78)	455 (365)
Diaphragm (ng/g)	6664 (3752)	4557 (697)	4396 (2743)	4306 (1120)	3169 (156)	4465 (2532)	1294 (490)	787 (435)	831 (302)	813 (151)	917 (304)	915 (96)	2163 (666)	2214 (1421)	2457 (1357)	2169 (381)	2348 (1174)	2659 (1435)

0.03, respectively). In the brain, atenolol concentrations tended to increase, whereas metoprolol and propranolol remained stable. Concentrations in bile were difficult to interpret because of the absence of measurement at 12 h. However, there was a trend to decrease until 12 h, which was greater for propranolol than for the other two BB. Concentrations in the diaphragm and iliopsoas muscle remained stable over time, but were significantly higher in the diaphragm than in the iliopsoas muscle for the three BB at all sampling times ($p<0.001$ for all three).

Discussion

Concentrations at 0 h post-mortem confirm that in vivo distribution of these three BB is strongly influenced by their lipophilicity, since for a similar pK_a, propranolol (K_p of 5.39) accumulates in cardiac muscle, lungs, brain, iliopsoas muscle and diaphragm, metoprolol (K_p of 0.18) only shows accumulation in the lung apexes, and atenolol (K_p of 0.01) is homogeneously distributed in the various sites (i.e. accumulates in none). These results suggest that K_p may be a better indicator of tissue distribution than V_d since propranolol, with a similar V_d and a higher K_p than metoprolol, seems to accumulate more intensely in tissue than the latter.

In cardiac muscle, the increase in concentrations of propranolol and metoprolol over time is likely due to redistribution from the lungs and/or stomach content. Redistribution from the lungs is suggested by a significant decrease in the levels of these BB in the four lung sites. This phenomenon, reported for lipophilic molecules, could occur either along a concentration gradient or via the lung vessels [4, 5, 13]. Based on concentration diminution, redistribution would be more intense from the lung bases than from the apexes, as previously described [6]. The decline of propranolol concentrations in the stomach content after 6 h also suggests redistribution from this site towards cardiac muscle because of the close anatomical relationship between the latter and the stomach [2–4].

For cardiac blood, the early decrease of the three BB must be discussed. To our knowledge, redistribution from cardiac blood towards the surrounding tissues has not been described. On the other hand, the possibility of agonal flow of cardiac blood towards the large vessels has already been raised in man after attempted resuscitation [7, 27] and in animals after killing them using potassium chloride [28], which induces ventricular fibrillation with cardiac arrest in diastole. However, it was not observed after the animals were killed using thiopental [29]. The secondary increase in both the left and right cardiac blood concentrations probably corresponds to classic redistribution of basic lipophilic molecules from lung parenchyma and/or cardiac muscle and/or stomach content [30]. Diffusion mechanisms from the last two tissues mentioned are certainly predominant here as myocardial tissue concentrations are higher than those in cardiac blood [31]. At last, the fact that these variations were more marked for propranolol than for the other two BB confirms the influence of lipophilicity on these phenomena of redistribution.

In peripheral blood, interpretation of the concentrations must take into account the following: the high standard deviation values essentially linked to the difficulty of drawing peripheral blood at late post-mortem intervals, on the one hand; that samples were not taken from the femoral vein, but from the inferior vena cava (once again because of the small quantities of blood available), which implies a reasoning in terms of anatomical relationships that is not necessarily the same as that of the reference studies, on the other. As was mentioned above, the most likely hypothesis to explain the elevated concentrations in peripheral blood from 2 to 12 h is the agonal flow of right cardiac blood. The siphon effect during sampling, sometimes suggested in the literature [28], seems very unlikely here as the inferior vena cava was clamped before sampling. The hypothesis of redistribution from the liver parenchyma [32] is also improbable here owing to the relative stability of liver parenchyma concentrations. Moreover, the concomitant decrease in bile concentrations may have contributed to the increased concentrations in peripheral venous blood, but the absence of bile measurement at 12 h restricts its appreciation. It thus appears that the variations of peripheral blood concentrations are not directly proportional to lipophilicity since propranolol seems to change less markedly than atenolol and metoprolol. Lastly, the fact that peripheral blood concentrations show non-negligible variations means that great care should be taken when interpreting post-mortem blood concentrations.

The presence of the three BB in the stomach content at 0 h post-mortem, although the drugs were administered by injection, can be explained by ante-mortem distribution. The very early increase in propranolol and metoprolol concentrations is probably partly due to redistribution from the liver parenchyma by transparietal diffusion, and partly by bile release in the duodenum and regurgitation to the stomach. The subsequent decrease in propranolol and metoprolol gastric concentrations could contribute to higher levels in the liver and/or cardiac muscle [1, 3, 7]. Lastly, the progressive increase in atenolol gastric concentrations could reflect redistribution from the liver [32]. The molecules would thus behave differently according to their lipophilicity.

In the liver, the slight increase in propranolol and metoprolol concentrations over time could be due to redistribution from the stomach [1] or the gall bladder [33]. The hypothesis of continued liver metabolism in the early post-mortem period, as suggested for other molecules [34, 35], seems rather unlikely here, as it would lead to decreased concentrations of the parent molecules.

Interpretation of bile concentrations is difficult because of the absence of measurement at 12 h. Atenolol, a hydrosoluble molecule, presented high bile concentrations at 0 h post-mortem, whereas propranolol, which undergoes enterohepatic cycling in man, was much less concentrated in bile than the other two BB. These changes likely reflect redistribution towards adjacent anatomical structures, especially the liver and inferior vena cava.

In urine, the variations observed are unquestionably related to lipophilicity since atenolol concentration was

stable over time, while propranolol and metoprolol varied according to the post-mortem intervals. Moreover, the ion-trapping phenomenon could play a part in the accumulation of these basic compounds in this acidic environment during the first 48 h [12].

Concentrations in the two muscular compartments remained stable over time, as described for other substances [36, 37], but the concentrations in the diaphragm were significantly higher than those in the iliopsoas muscle at all sampling times for the three BB. These differences could arise either from the fact that the blood flow to the diaphragm is greater than that to the peripheral muscles, or from redistribution from the lungs, stomach content or liver towards the diaphragm [1, 7, 32]. Here, a redistribution mechanism (probably from lung bases) depending on lipophilicity is suggested by the tendency of propranolol concentrations to increase in the diaphragm, while atenolol and metoprolol levels became stable after 2 h post-mortem.

In the vitreous humour, metoprolol and atenolol concentrations increased significantly while propranolol concentrations rose only slightly, suggesting a negative relation with lipophilicity at this site, as previously described [24]. *A contrario*, the absence of post-mortem redistribution of ethanol in the vitreous humour does not support the hypothesis of preferential redistribution of more hydrosoluble molecules [38]. The hypothesis of redistribution via the airways and then the nasopharynx, skull base and eye globe, which has been proposed for liposoluble molecules, such as MDMA, as well as for ethanol [37], cannot explain the difference of behaviour between the three BB.

In the brain, atenolol concentrations increased significantly whereas metoprolol and propranolol concentrations remained stable. In vivo, only a very small fraction of the hydrosoluble atenolol crosses the blood–brain barrier, whereas metoprolol and propranolol, which are liposoluble, cross it readily [10, 22], as confirmed by the relative concentrations of the three BB in the brain at 0 h post-mortem. This is consistent with the finding that post-mortem diffusion into the brain only concerned the most hydrosoluble molecule. Finally, although the brain could be used to estimate the post-mortem interval [39], the increase in concentration of the three BB indicates that this matrix would not represent an interesting alternative matrix for post-mortem BB measurement.

Conclusion

In conclusion, this study confirms that the lipophilicity of a molecule is not the only predictive factor of its post-mortem redistribution and suggests that this parameter must be considered according to the anatomical sampling site. While it clearly appears that molecular lipophilicity influences post-mortem redistribution in the thoracic–abdominal compartment consisting of the stomach, lungs, cardiac muscle, cardiac blood, liver and diaphragm, its contribution is much less clear in other sectors and tissues such as peripheral blood, skeletal muscle, brain and vitreous humour. Moreover, the finding that propranolol undergoes larger variations than metoprolol (which has a lower partition coefficient but a similar volume of distribution) in the thoracic–abdominal compartment suggests that drug redistribution would be more influenced by the partition coefficient than by the volume of distribution. Indeed, these results lead us to reconsider the generally accepted notion that post-mortem redistribution mainly concerns the molecules with a V_d of more than 3 L/kg [16]. Lastly and more generally, they underline the difficulty of interpreting these redistribution phenomena.

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