

Cocaine-Induced Dopamine Overflow Within the Nucleus Accumbens Measured by In Vivo Microdialysis: A Meta-Analysis

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KEY WORDS microdialysis; DA overflow; nucleus accumbens; shell and core; cocaine; meta-analysis

ABSTRACT A meta-analysis was conducted on data obtained from published articles which used in vivo microdialysis to assess dose-response curves of cocaine on dopamine (DA) overflow within the nucleus accumbens (NAC). Different experimental and biological parameters such as route of administration (ip, sc, iv, local), rat and mouse strains, gender, age aspects, and regions cannulated (NAC core and shell) were considered. Data from 116 experiments involving 833 animals (out of 266 publications) fulfilled our selection criteria and were analyzed in relation to absolute basal DA levels, the maximum peak of DA overflow (peak [%] baseline) and the time when this peak (peak time) occurred. Our meta-analysis revealed that absolute basal DA levels lie at 2.39 nM (median of all experiments) and that cocaine-induced DA overflow in the NAC is significantly enhanced in a linear dose-response fashion within the applied dose range as the regression function increases following either iv or ip administration. Peak time was reached fastest in iv experiments and slowest following local application. Furthermore, it was shown in ip experiments that the higher the dose, the longer it took to reach the zenith. Results from the NAC shell region displayed greater DA overflow as compared with the NAC core. DA overflow properties following cocaine treatment in mice did not differ from that in rats. Thus, neither species differences nor other biological factors such as, age, gender, and rat/mouse strain have a pronounced impact on cocaine-induced DA overflow. Technical parameters of the microdialysis procedure such as calcium concentration of the perfusion medium and collected sample amount have also no significant effect in terms of DA overflow properties (peak [%] baseline and peak time) following cocaine treatment. In conclusion, these data may be deemed useful for textbook knowledge and a better comparability of data given by the generalization of already existing data as well as for investigators in maximizing the effect of cocaine-induced DA overflow. Finally, this study exemplifies how meta-analyses may be applied to a wide range of data within the field of neurochemistry. **Synapse 62:243–252, 2008.** © 2008 Wiley-Liss, Inc.

INTRODUCTION

In vivo microdialysis has been used to quantitatively study the chemical composition of interstitial tissue fluid. Virtually, every soluble molecule in the interstitial space fluid can be measured by microdialysis. This method has been applied to a variety of species either under anaesthetized or freely moving conditions and even in situ measurements in humans. In vivo microdialysis allows the observation of neurotransmitter release in specific brain nuclei, while a systemic or local administration of a certain drug is

conducted. Behavioral observations can be made in parallel and neurochemical events may then be related to the behavioral responses.

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Numerous microdialysis studies concern themselves with the mesolimbic DArgic system, since it is known that dopamine (DA) has a gating function in reinforcing stimuli such as food, sex, and drugs of abuse (Spanagel and Weiss, 1999). In particular, the administration of a drug of abuse e.g. cocaine leads to an overflow of DA within the nucleus accumbens (NAC). In the literature, one can find a host of studies assessing the effects of cocaine on DA overflow within the NAC, as it is known that cocaine directly interacts with the DA transporter and produces a pronounced DA peak in the NAC. Although hundreds of studies have been published in this regard—and all of them clearly demonstrate that DA overflow is enhanced following cocaine administration—relevant questions are difficult to resolve. (i) What does the dose-response relationship look like—is the correlation linear or does it level down to a plateau in the so far studied dose range? (ii) Does the route of administration influence peak magnitude and time? (iii) Are there species, strain, and/or sex differences in the dose-response relationship? (iv) Is there a difference between the NAC core and shell region with regard to the properties of cocaine in inducing DA overflow? (v) Do technical parameters of the microdialysis set-up such as the calcium concentration in the perfusion fluid or the sample amount influence cocaine-induced DA overflow? It would be extremely demanding to design a set of experiments that would give us adequate answers to these general and complex questions. However, the use of meta-analysis of existing published data represents a powerful tool in dealing with these questions.

The idea of meta-analysis goes back to 1977, when the first meta-analysis on clinical data was obtained (Smith and Glass, 1977). The term “meta-analysis” itself was introduced by Glass (1976). It is defined as a summary of primary studies, working with quantitative as well as statistical methods. The purpose of using meta-analysis is to integrate the findings of a large collection of individual studies and it represents a method for data analysis, in addition to come up with universally valid statements on a certain topic. Further, most of the earlier meta-analyses were estimated on clinical data. Only in recent years were a few meta-analyses performed in basic and preclinical animal research. These meta-analyses concerned themselves mainly with behavioral measures such as conditioned place preference (Bardo et al., 1995) or learning (Jonasson, 2005). Here, we investigated and collected the data of the existing literature on the effect of cocaine on DA overflow within the NAC. With this approach a generalization of the findings is possible. Thus, a meta-analysis approach in neurochemistry would allow us to draw general conclusions with regard to textbook knowledge, guide researchers designing their future experiments in a more appropriate

way and help reduce the number of animals required for a specific experiment.

METHODS

Literature search and coding of variables

A literature search was performed on pubmed (<http://www.ncbi.nlm.nih.gov/entrez/query.fcgi>). The search included all the following keywords simultaneously: cocaine, DA, microdialysis, accumbens. No particular journals were prechosen. A total of 266 entries were found. The first objective was to include every single publication that was found reporting on this certain topic in order to get a general summary. In the meta-analysis, only publications (named in the references) were included, where indeed a microdialysis experiment was conducted and peak (%) baseline and time were estimated. Peak (%) baseline was defined as the highest percentage deviation from the baseline value, set at 100%. Peak time was defined as the time, when the highest percentage deviation occurred. Data sets were excluded that did not mention a parameter that was necessary for data analysis (mean, SEM, *n*).

The following variables were extracted from the publications and used for further analysis:

- i. Rat or mouse strain, sex, and age (weight) of the animals.
- ii. Coordinates of probe placement according to the stereotaxic atlas of either Paxinos and Watson (several years), Pellegrino et al. (1979), Franklin and Paxinos (1997), or König and Klippel (1974).
- iii. Doses of cocaine applied, as well as route of administration (ip, iv, sc or local).
- iv. Sample time of microdialysates.
- v. Peak (%) baseline (= mean value of peak [%] baseline over the animals of each experiment) and the time when the maximum peak occurred (= mean value of the time over the animals of each experiment) as well as the number of animals used (it should be noted that it was easy to extract the variable “peak (%) baseline,” due to a common presentation of data in microdialysis studies). To obtain the values of peak (%) baseline, measurements were taken directly from the appropriate graph.
- vi. Absolute basal DA levels. As these values were presented in different units, they were then recalculated to molarity (nM).
- vii. Calcium concentration in the Ringer solution or artificial CSF (mM).
- viii. Length of the probe membrane (mm).
- ix. Perfusion rate ($\mu\text{l}/\text{min}$) and sample time (min) as well as the factor from both (collected sample amount).

Data analysis

Normally, the observation unit of a meta-analysis is not the observation unit of one single subexperiment but the experiment in total. Thus, the variables of the meta-analysis are not values that are collected from a single animal but means, percentages, numbers etc. that are related to the entire experiment.

The main objective of the data analysis is the variable “peak (%) baseline” (= mean value of peak [%] baseline of each experiment), with one value for each experiment. For a graphical representation of peak (%) baseline, we used forest-plots. A forest-plot is a scattergram of the variables “experiment” and “peak (%) baseline” to which the confidence interval of peak (%) baseline is added for each experiment. In the two forest-plots (see section results), the experiments are ordered by the variable “year of publication.” We used weighted means to combine peak (%) baseline over groups of experiments (e.g. over the experiments with route of administration ip and iv). The weight of peak (%) baseline of a specified experiment is proportional to the inverse variance of peak (%) baseline in this specified experiment. The same weights are used for additional purposes: e.g. for the computation of weighted correlations, for weighted analysis of variance and for the estimation of regression functions.

In one part of the data analysis, peak (%) baseline is regarded as a linear function of the cocaine administered: since a greater amount of cocaine increases more DA than a smaller one, f is assumed to be increasing. Further we checked, whether the slope of f is positive. The meta regression analysis was performed as proposed by Hedges and Olkin (1985).

RESULTS

Out of 266 publications, 66 articles with a total of 143 experiments fulfilled our selection criteria. However, only 81.8% of the experiments (= 116 experiments) revealed a complete data set and, which were then used for further analysis. Only a few publications (4%) emerged before 1990. 47% of all articles were obtained after the year 2000.

In Table I, the number and percentage of experiments with respect to different routes of administration are shown. Almost 80% of the investigators chose the ip route of administration. Thus, cocaine was administered five times more ip than iv. A minor percentage of the investigators administered the drug sc or locally (direct infusion of cocaine into the NAC through the microdialysis probe). Over 90% of the experimenters worked with rats, the minor percentage worked with mice (Table I). The number of animals in each experiment ranged from 3 to 18. The mean is about 7 and the median 6.5. In 25% of the experiments, five or less animals were used and in

TABLE I. Number and percentage of experiments with different routes of administration; number and percentage of experiments with different species; number of animals used per experiment; total sum of all animals used in respect to the route of administration

Variable	N	%
Route of administration		
ip	91	78.5
iv	17	14.7
Local	3	2.6
sc	5	4.3
Total number of experiments	116	100
Species		
Rat	108	93.1
Mouse	8	6.9
Animals used per experiment (n)		
Mean	7.2	
Median	6.5	
Quantile 75%	8.0	
Quantile 25%	5.0	
Max	18	
Min	3	
Sum of n		
All	833	
ip	673	80.8
iv	99	11.8
Local	21	2.5
sc	40	4.8

25% of the experiments eight or more animals were included in the studies. The total number of used animals in all experiments was 833 (Table I).

A forest-plot (Fig. 1A) displays the values for the variable “peak (%) baseline” in the 91 experiments with the ip route of administration. Note that row 1 in this diagram refers to the weighted mean and its confidence interval. These 91 experiments are ordered according to the year of publication. The median of the peak (%) baseline is 333, the 75% quantile and the 25% quantile of peak (%) baseline are 430 and 240, respectively. That means that in 25% of the experiments, peak (%) baseline levels are 430 or greater, whereas in 25% of the experiments they are 240 or lower. The weighted mean is 297.7, with a confidence interval of 294.0–301.4. Over the years, no large variability was observed, except in one experiment where peak (%) baseline reached 1500% with 30 mg/kg, which is uncommon. In another 17 experiments, the route of administration was iv (Fig. 1B). The weighted mean in these experiments is 243.3 with a confidence interval of 232.1–254.5. The mean values for ip and iv administrations are quite similar compared with local and sc treated subjects. Thus, comparing the different routes of administration, peak (%) baseline is significantly different ($P \leq 0.01$). In Table II, peak (%) baseline and peak time are given with respect to the route of administration. Although the average administered dose in ip experiments is more than eight times higher than the average dose in iv experiments, the peak (%) baseline levels are comparable in ip and iv experiments. The mean dose in the ip administered experiments is greater than the median, which shows a deviation to the right. The minimum dose was 3 mg/kg and the maximum 40 mg/kg. Within the 91 experi-

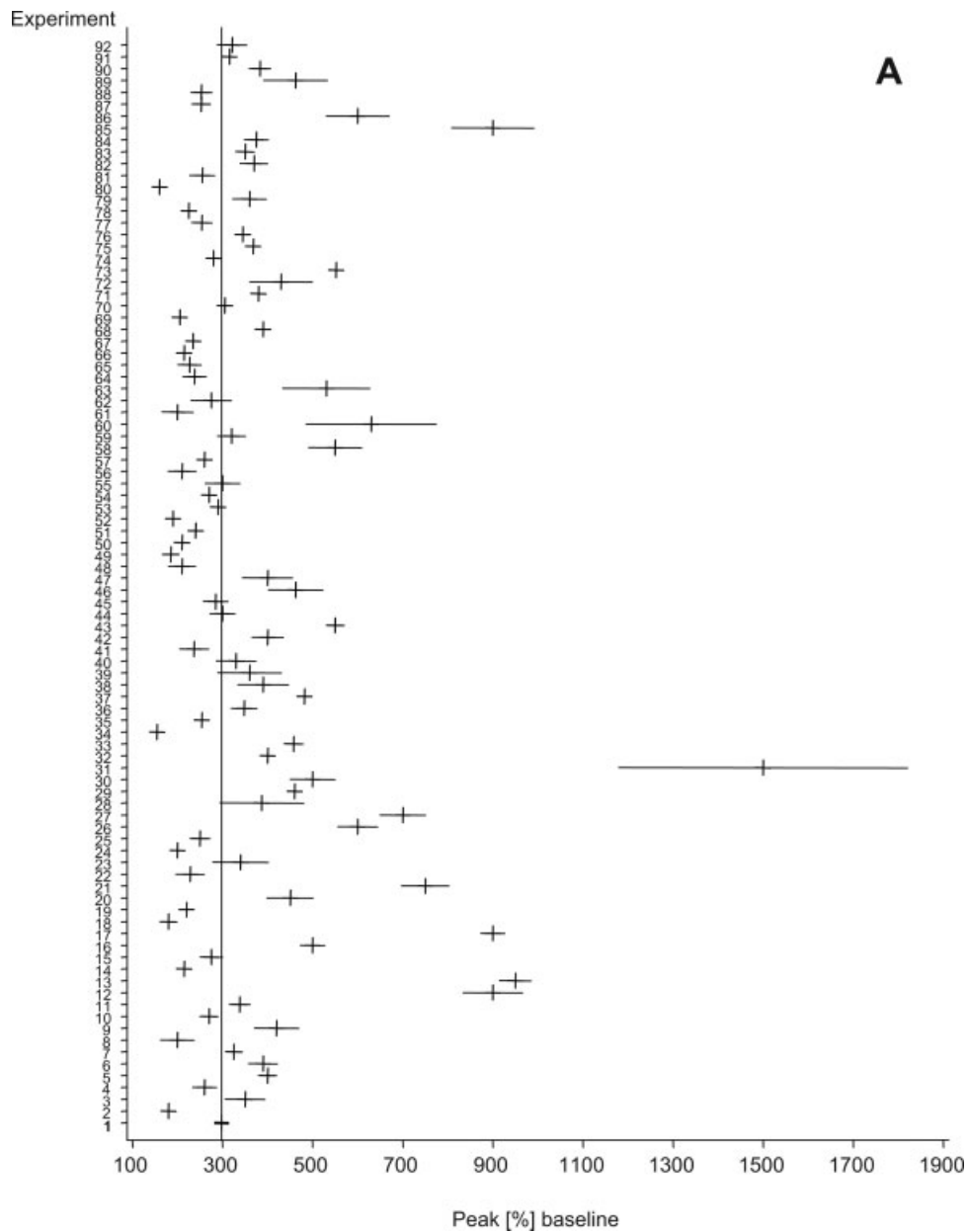


Fig. 1. Forest-plots of the variable “peak (%) baseline” measured in 91 (ip; **A**) or 17 (iv; **B**) experiments, respectively. Experiments sorted by year of publication. Rows 2–92: contain the value and confidence interval of peak (%) baseline, the row “1” shows the weighted mean and its confidence interval. The vertical line extends the weighted mean in order to compare the extracted data. (A) 2 Brown et al., 1991; 3, 4 Hooks et al., 1991; 5 Maisonneuve and Glick, 1992; 6, 7 Hooks et al., 1992; 8, 9 Steketee, 1993; 10 Kimura et al., 1993; 11 McNeish et al., 1993; 12, 13 Maisonneuve et al., 1994; 14–21 Camp et al., 1994; 22 Nation and Burkey, 1994; 23 Essman et al., 1994; 24–26 Hemby et al., 1995; 27, 28 Kankaanpää et al., 1996; 29 Reith et al., 1997; 30, 31 Pierce et al., 1997; 32 Morgan et al., 1997a; 33 Morgan et al., 1997b; 34–36 Parsons et al., 1998 and 1999; 37 Morgan and Dewey, 1998; 38 Kuczynski and Segal, 1999; 39 Tolliver et al., 1999; 40 Parson et al., 1999; 41

Hedou et al., 1999; 42 Beyer and Steketee, 2000; 43 Gerasimov et al., 2000; 44–46 Andrews and Lucki, 2001; 47 Lindholm et al., 2001; 48, 49 Alvarez Fischer et al., 2001; 50–57 Mikkola et al., 2001; 58, 59 Rocha et al., 2002; 60 Kankaanpää et al., 2002; 61–63 Steketee and Goeders, 2002; 64, 65 Müller et al., 2002; 66–71 Cadoni et al., 2003; 72 Leri et al., 2003; 73 Schiffer et al., 2003a; 74 Lu et al., 2003; 75, 76 Bubar et al., 2003; 77 Shimosato et al., 2003; 78–81 Zocchi et al., 2003; 82 Fadda et al., 2003; 83 Schiffer et al., 2003b; 84–86 O’Dell and Parsons, 2004; 87, 88 Lodge and Grace, 2005; 89 Andrews et al., 2005; 90–92 Gurkovskaya et al., 2005. (B) 2 Bradberry and Roth, 1989; 3, 4 Moghaddam and Bunney, 1989; 5 Bradberry et al., 1993; 6, 7 Baptista et al., 1993; 8–12 Baumann et al., 1994; 13–15 Pontieri et al., 1995; 16 Rothman et al., 1996; 17 Duvauchelle et al., 2000; 18 Pepper et al., 2001.

ments, 25% were performed with a dose lower than 10 mg/kg and a dose higher than 20 mg/kg, respectively. The most common doses in the ip experiments

were 10 mg/kg (35.2% of all ip experiments), 15 mg/kg (18.7%), and 20 mg/kg (16.5%). The mean dose in the iv administered experiments is identical with the

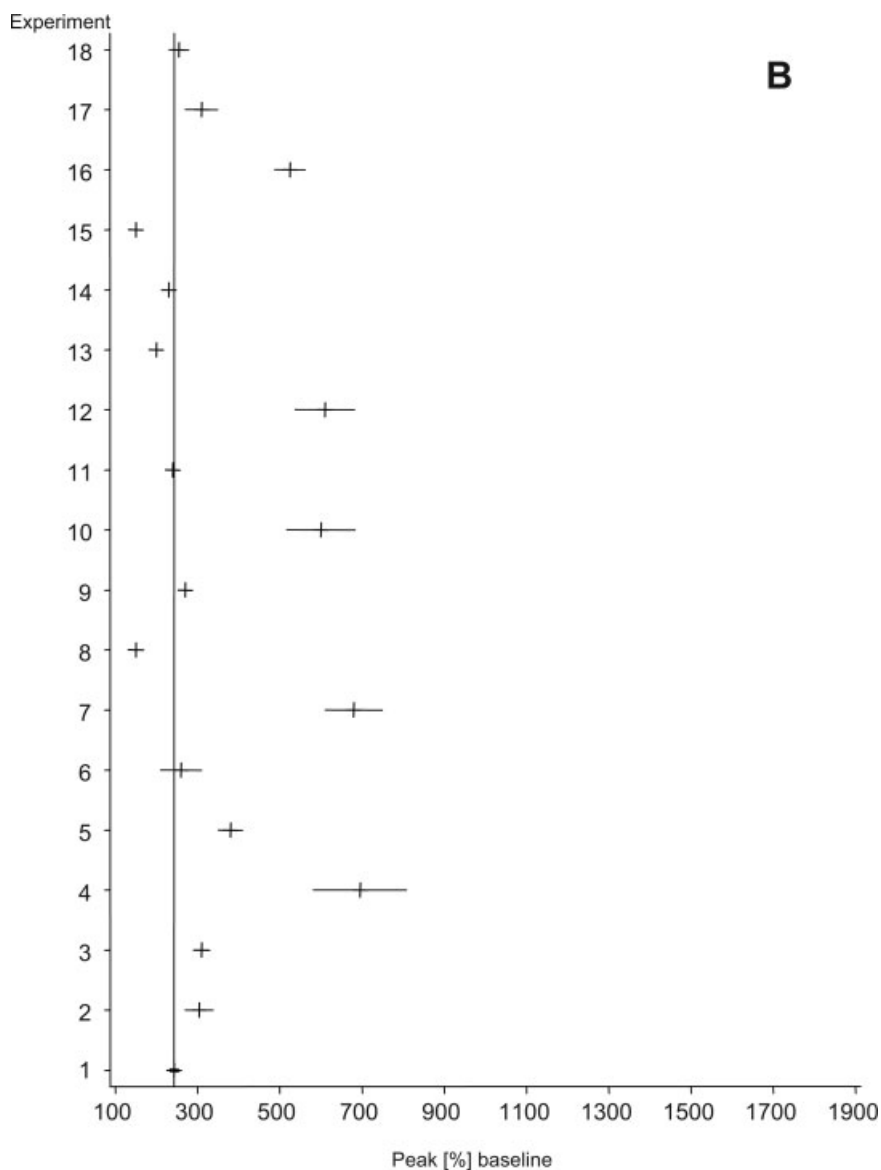


Figure 1 (Continued.)

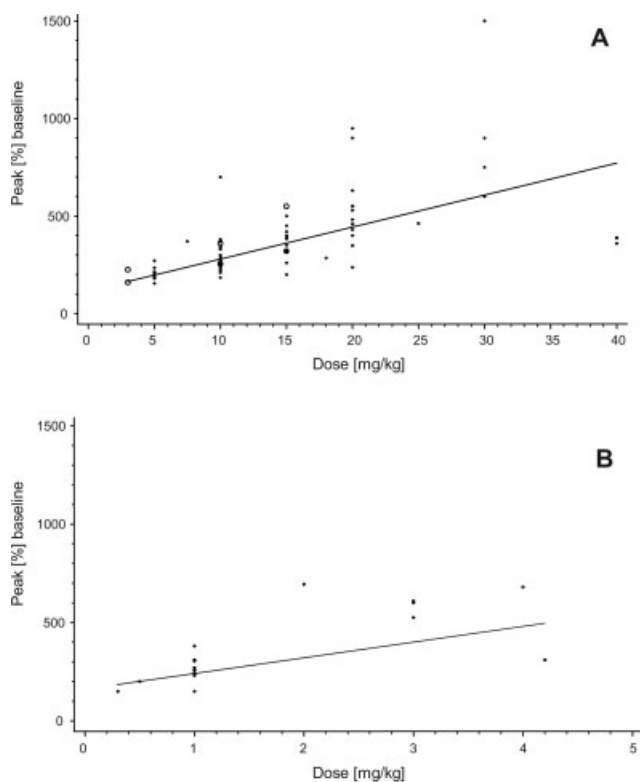
median. The weighted correlation between dose and peak (%) baseline for 91 experiments that used ip as the route of administration was $r = 0.71$ ($P = 0.001$). The correlation between the amount of applied cocaine and released DA is also highly significant. The correlation is positive i.e. in experiments that used higher doses, higher amounts of DA were released. Figure 2A contains a detailed illustration of this correlation. It displays a scatter plot of the variables “dose” and “peak (%) baseline.” We fit a straight line to the data and got a regression coefficient of 16.4 (CI: [15.8; 17.1]). That means an increase in the dose by 1 mg/kg leads to an increase of peak (%) baseline of 16.4 percentage points. The statistical spread in the ip experiments increased with higher doses (Fig. 2A). The regression function for rats alone is identical with the

function rats + mice (data not shown). The weighted correlation between dose and peak (%) baseline for the 17 experiments that used the iv route of administration, is 0.66 ($P \leq 0.01$). The correlation between the administered cocaine and the released DA is also significant and positive. Figure 2B contains the scatter plot for the variable “dose” and “peak (%) baseline” for the experiments, which used the iv route of administration and the regression function was fit to the cloud of dots. We fit a straight line to the data and got a regression coefficient of 80.0 (CI: [62.0; 98.1]). That means an increase in the dose by 1 mg/kg leads to an increase of peak (%) baseline of 80.0 percentage points.

Regarding the interpretation of the peak time, one has to mention that the time window of measuring

TABLE II. Characterisation of dose, peak time and “peak (%) baseline” according to the route of administration (not weighted)

Variable		ip	iv	Local	sc
Dose (mg/kg) or (mM) for Local	Mean	14.5	1.7	4.4	4.4
	Median	10.0	1.7	3.0	3.0
	Quantile 75%	20.0	3.0	10.0	5.0
	Quantile 25%	10.0	1.0	0.3	3.0
	Max	40.0	4.2	10.0	10.0
	Min	3.0	0.3	0.1	1.0
Peak time (min)	Mean	32.9	17.6	120.0	74.0
	Median	30.0	20.0	140.0	70.0
	Quantile 75%	40.0	20.0	140.0	90.0
	Quantile 25%	20.0	20.0	80.0	60.0
	Max	80.0	20.0	140.0	90.0
	Min	10.0	10.0	80.0	60.0
Peak (%) baseline	Mean	377.2	362.9	722.0	228.2
	Median	330.0	304.0	826.0	220.0
	Quantile 75%	430.0	525.0	835.0	286.0
	Quantile 25%	240.0	240.0	505.0	150.0
	Max	1500.0	695.0	835.0	335.0
	Min	155.0	150.0	505.0	150.0
	Mean (weighted)	297.7	243.3	646.8	196.7

Fig. 2. Scatter plot of “peak (%) baseline” and dose (experiments with rats: dots, experiments with mice: circles) and regression line in 91 (ip; **A**) or 17 (iv; **B**) experiments, respectively.

DA in the microdialysis samples mostly varied from 10 to 20 min. As expected, according to the route of administration there are differences in peak time (Fig. 3). Thus, the shortest time to reach the maximum peak was observed following iv administration whereas the longest time to reach the maximum peak occurred following local application. The higher the dose one is using in ip experiments, the longer it takes to reach the maximum peak (data not shown).

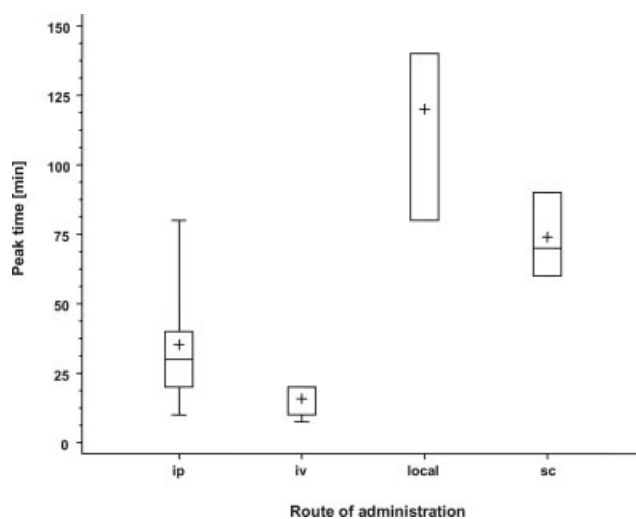


Fig. 3. Box plot of peak time in respect to route of administration.

Thus, a highly significant correlation of dose with peak time could be found in ip experiments ($r = 0.44$; $P \leq 0.001$). Neither iv nor local or sc experiments reached significance with respect to dose and peak time correlation.

The NAC is a heterogenous brain area, which can be mainly divided into the core and shell region. Only in 22 experiments were these region specific differences taken into account. In Figure 4A, peak (%) baseline was plotted for the different regions cannulated (NAC, NAC shell, NAC core). The slope of the dose-response-curve is bigger in the NAC shell than in the core region or the NAC in general ($P \leq 0.05$).

In rats, the mean basal DA levels in the NAC were 7.88 nM but varied between 124 and 0.03 nM (Table III). The median was 2.58 nM. 25% of the publications found basal DA levels lower than 1.31 nM and 25% higher basal DA levels than 4.25 nM. In comparison,

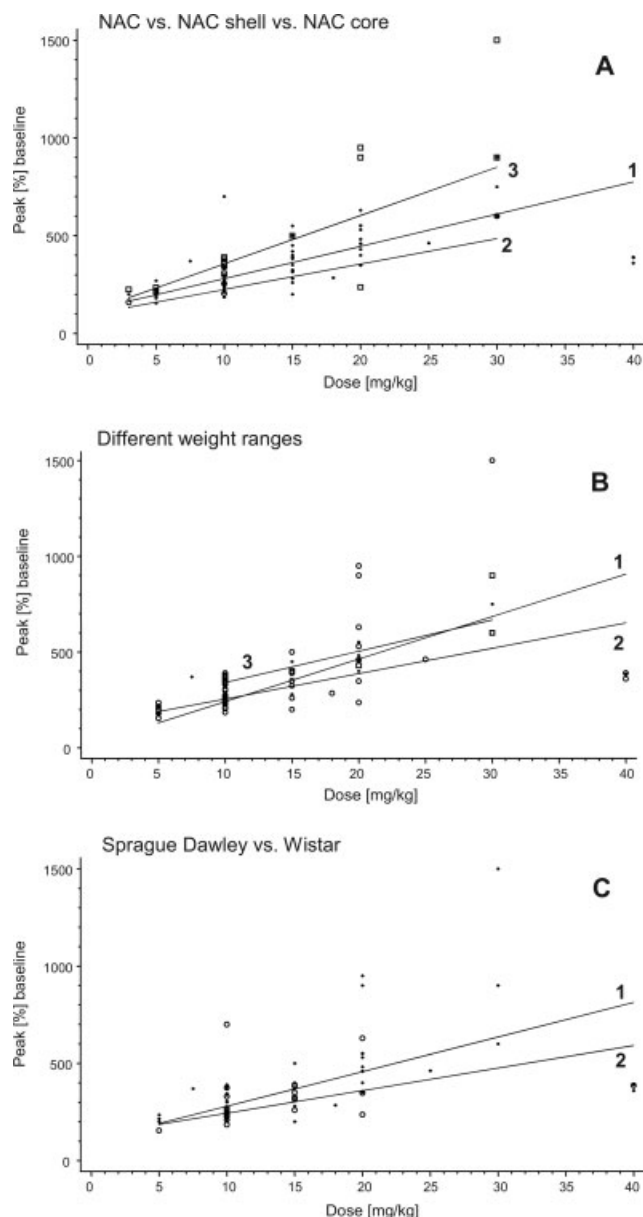


Fig. 4. **A:** Scatter plot of “peak (%) baseline” and dose; ip experiments within the NAC (subregion not defined) (dots), ip experiments within NAC core region (open circles), and ip experiments within NAC shell region (open squares). Regression line: 1, NAC; 2, NAC core; 3, NAC shell. **B:** Scatter plot of the variables “peak (%) baseline” and “dose,” ip experiments within different ages given by the weight in rats. Ranges and regression curve: 1, 187–255 g; 2, 255–322 g; 3, 322–390 g. **C:** Scatter plot of “peak (%) baseline” and dose; ip experiments with Sprague Dawley rats (dots) and ip experiments with Wistar rats (open circles). Regression line: 1, Sprague Dawley; 2, Wistar.

the mean basal DA of mice was 2.25 nM but varied between 3.8 and 0.90 nM (Table III). The median was 2.15 nM. 25% of the publications found basal DA levels lower than 1.15 nM and 25% basal DA levels higher than 3.35 nM. Finally, comparing the basal DA levels of rats in the NAC, NAC core and shell revealed no apparent differences between the regions (Table III).

TABLE III. Basal DA (nM) in rats and mice NAC; basal DA in rats itemized into NAC (n = 38), NAC core (n = 2), and NAC shell (n = 9)

	Mice	Rats	NAC	NAC core	NAC shell
Mean	2.25	7.88	9.24	4.15	2.95
Median	2.15	2.58	2.39	4.15	2.75
Max	3.8	124.04	124.04	4.25	6.4
Min	0.90	0.03	0.03	4.05	0.63
Q1	1.15	1.31	1.31	4.05	0.74
Q3	3.35	4.25	4.58	4.25	4.15

In addition, several other biological parameters such as gender, age (estimated by body weight), and rat/mouse strain were analyzed. 87.9% of the experiments used male rats. In the other experiments female rats or both genders were used. In some studies, the gender could not be extracted from the Methods section and thus the total number of experiments including female animals is too low to draw any general conclusion (data not shown). The age of the rats per experiment was mainly measured by weight (g) (in 85.2% of all experiments). The mean weight range was between 187 and 390 g. The mean weight was 289.9 g. Rats were divided into three age groups (group 1, 187–255 g; group 2, 255–322 g; group 3, 322–390 g). Figure 4B, shows that the slope of the dose-response-curve in group 1 was higher than in group 2 ($P \leq 0.05$). Seven different rat and mouse strains were used. However, in most of the experiments, Sprague Dawley rats (53.5%) and Wistar rats (19.8%) were used. A strain comparison between Sprague Dawley and Wistar rats (most commonly used rat strains) revealed no differences in peak (%) baseline (Fig. 4C). Slope comparison between both curves showed no significant difference.

Technical parameters of the microdialysis set-up were analyzed as well. The calcium concentration of the perfusion medium ranged between 1.1 and 3.4 mM. In 45.5% of the experiments a calcium concentration of 1.2 mM was used. For comparison, three groups were generated and statistically compared. Between group 1 (1.1–1.22 mM), group 2 (1.26–2 mM), and group 3 (2.2–3.4 mM) no significant differences could be found on cocaine-induced DA overflow properties (peak [%] baseline). The regression coefficient of group 1 was 16.1 (CI: [15.4, 18.5]), of group 2 17.4 (CI: [11.6, 23.1]), and of group 3 16.5 (CI: [14.8, 18.1]).

The length of the probe membrane ranged from 1 to 3, whereas in 78% of the experiments a 2-mm membrane was used. Group comparisons (1 mm vs. 2 mm vs. 3 mm) did not yield any significant differences in terms of DA overflow properties (peak [%] baseline and peak time) following cocaine treatment.

Sample time ranged between 10 and 40 min—in 56% of the experiments the samples were collected for 20 min—and the perfusion rate ranged between 0.25 and 5 μ l/min. The hereof combined factor “collected sample amount” ranged between 5 and 100 μ l

and was further statistically analyzed. Between generated groups (group 1, 5–15 μl , group 2, 15.6–38 μl , group 3, 40–100 μl) no significant differences could be found in respect to cocaine-induced DA overflow. The regression coefficient of group 1 was 14.3 (CI: [12.9, 15.7]), of group 2, 14.1 (CI: [12.9, 15.4]), and of group 3, 19.0 (CI: [14.8, 20.3]).

DISCUSSION

In vivo microdialysis represents an ideal tool for studying the effects of drugs on neurotransmitter release. For an effective planning of microdialysis experiments, it would be very helpful to follow some guidelines that define technical, experimental, and biological parameters. Technical parameters for in vivo microdialysis experiments and adjunct neurochemical analyses are usually well defined and as shown here have no major impact in terms of DA overflow properties (peak [%] baseline and peak time) following cocaine treatment. However, experimental and biological parameters such as the number of animals per experiment, species and animal strains, gender, age, brain area, doses of applied drugs, time windows of measurement can largely vary between studies and it would be appropriate to define these parameters more precisely. This can be accomplished by a meta-analysis. The concept of a meta-analysis as used in the present study is novel in the field of neurochemistry and has so far only been applied to behavioral and toxicological basic research experiments (Bardo et al., 1995; Jonasson, 2005).

Our meta-analysis of studies on cocaine-induced DA overflow in the NAC shows that the experimenters mainly used rats—Sprague Dawley and Wistar were the most common strains—and administered cocaine ip. This choice is very practical as rats are easy to handle and transferable in an awake state to the microdialysis system without inducing undue intense stress to the animals. This can be also done by a not too experienced experimenter including the ip injection. For good statistical analysis, it is essential to choose a reasonable number of animals. The herein estimated mean of 7 reflects thoroughly planned experiments.

In microdialysis studies, it is a universally valid agreement to express the data relative to the (%) baseline. However, in some studies absolute values of DA concentrations in the extracellular space are given as well. In the present study, we calculated for the mean basal DA levels in the NAC within the rat brain a concentration of 7.88 nM which varied between 0.03 and 124 nM. These large variations in baseline values are due to the time of measurement, length of the probe membrane, individual differences, the exact placement, and miscalculations. It should be noted that microdialysis studies most likely pro-

vide an underestimation of the absolute dopamine concentration in the nanomolar range whereas recent voltammetric studies suggest that the concentrations may be considerably higher, perhaps in the micromolar range (Michael et al., 2005). Given these large variations in most studies data are expressed relative to the (%) baseline. The baseline values are usually the first three collected samples free of drug treatment. This baseline is set at 100% and the following values are calculated relative to that baseline. The peak represents the sample with the major content of DA.

Analysis of the route of administration with respect to peak (%) baseline revealed an increasing regression function for ip and iv injections with an optimal dose of 10 and 15 mg/kg cocaine because the distribution around the regression function is lowest here. Interestingly, the values for mice and rats do not differ. Thus, values for mice lay within the cloud of values obtained from the rats. One remarkable outlier was found at a dose of 30 mg/kg causing an increase of up to 1500% baseline (Pierce et al., 1997). One can predict a methodological mistake. In the future, an experimenter would be able to compare the data with the collection here and decide, if the results lie within the normal range.

Analysis of peak time is a critical issue as sample time can differ between studies. Nevertheless, the herein obtained results are in agreement with the fact that iv administered cocaine reaches the brain most rapidly, whereas an sc injection builds up a deposit and is released into the bloodstream over a longer time resulting in a delayed peak time, also in comparison to an ip injection. Surprisingly, local administration of cocaine takes the most time to reach the maximum peak but the parameter peak (%) baseline is highest. Focusing on the different doses and the occurrence of peak, it is important to note that the higher the dose, the longer it takes to reach the peak maximum.

Cannulating the NAC means also to make a decision on which part of the NAC should be examined. Although it is pointed out in the literature that cocaine shows a selective effect in the shell at lower doses and a preferential one at higher doses (Pontieri et al., 1995) most of the experimenters did not separate between core and shell. However, in 22 experiments region specific differences were taken into account and with these experiments we performed a meta-analysis. By the divergent regression curves, we were able to confirm the observation by Pontieri et al. (1995) on a general basis. Thus, the slope of the dose-response-curve is bigger in the NAC shell than in the core region or the NAC. In behavioral sensitization, not assessed herein, the responsiveness of DA transmission in the NAC core is increased with no effect or even a decrease in responsiveness in the NAC shell.

These results led to the hypothesis that NAC shell DA responds to primary, unconditioned, unpredicted stimuli, whereas NAC core DA is generally activated by any relevant stimulus and undergoes sensitization (Di Chiara, 2002).

By assessing the influence of several biological parameters on cocaine-induced DA release properties, we found that neither species (rat vs. mouse) nor strain differences (Sprague Dawley vs. Wistar rats) had a major impact (which does not exclude the possibility that other species or strain differences might exist). Marked gender differences were also not observed. However, the factor age might influence DA overflow variables. Thus, we found a difference between the lightweight group and the middleweight group, with a higher slope in the dose response curve in group 1.

In conclusion, with our meta-analysis of cocaine-induced DA overflow into the NAC we were able to show that the amount of DA, which is released into the nucleus, increased with higher doses, with a higher variability around the regression function. We did not find any evidence for plateau attainment. To make more general conclusions on DA release properties following cocaine treatment a further comparison to voltammetric studies should be also performed as this technique challenges the relevance of data obtained by microdialysis on DA levels following the application of DA reuptake inhibitors (Borland et al., 2005).

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