Kathryn S. Kalasinsky,[1] Ph.D.; Thomas Z. Bosy,[1] Ph.D.; Gregory A. Schmunk,[2] M.D.; Lee Ang,[3] M.D.; Vernard Adams,[4] M.D.; Shashi B. Gore;[5] John Smialek,[6] M.D.; Yoshiaki Furukawa,[7] M.D.; Mark Guttman,[7] M.D.; and Stephen J. Kish,[7] Ph.D.

Regional Distribution of Cocaine in Postmortem Brain of Chronic Human Cocaine Users

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ABSTRACT: We measured concentrations of cocaine and its major metabolites (benzoylecgonine, ecgonine methylester, norcocaine, and cocaethylene) in 15 autopsied brain regions of 14 human chronic cocaine users. Only slight differences were observed in concentrations of cocaine and its metabolites amongst the examined brain areas. Although it is likely that some postmortem redistribution of the drug must have occurred, our data are consistent with the possibility that behaviorally relevant doses of cocaine are widely distributed throughout the brain of humans who use the drug on a chronic basis. Consideration should therefore be given to the possible pharmacological and toxicological actions of cocaine in both striatal and extra-striatal brain areas in human users of the drug.

KEYWORDS: forensic science, cocaine, benzoylecgonine, ecgonine methylester, norcocaine, cocaethylene, dopamine, caudate, putamen, brain

Investigations into the mechanisms of the actions of cocaine (e.g., euphoria, extrapyramidal disturbances) have primarily focused on the dopamine-rich areas of the striatum (caudate, putamen, and nucleus accumbens), the brain area containing high concentration of the dopamine transporter, which transports released dopamine back into the nerve ending. Occupancy by cocaine of about 50% of the striatal dopamine transporter binding sites appears to be necessary in order to perceive the euphoric effect of the drug in the human (1). Much less attention has been devoted to the potential actions of cocaine in extra-striatal dopamine transporter-poor brain areas (e.g., prefrontal cerebral cortex, hippocampus) that could explain, for example, the negative long-term effects of the drug on cognitive processes reported in some users (2–4). This may be related in part to the general perception that cocaine, which binds at high potency to the dopamine transporter (5,6), accumulates selectively in the striatal brain region. In this regard, both *in vivo* (7,8) and *in vitro* (9) data indicate that cocaine, at very low radiotracer doses, preferentially accumulates in those striatal regions of mammalian brain that contain high concentration of the dopamine transporter.

As cocaine abuse represents a significant public health issue worldwide, it is important to know whether, in human chronic users of the drug, the pharmacological and potentially neurotoxic effects of cocaine are likely to involve the entire brain or, conversely, be restricted to the striatal brain area. In principle, this question could be addressed to some extent in a human neuroimaging study employing radiolabeled cocaine. However, for ethical reasons, such an investigation cannot employ the high pharmacological, and sometimes lethal doses of cocaine that are self-administered by the cocaine user often in a "binge" pattern of use. An alternate strategy, the use of postmortem brain material from chronic cocaine users, is limited by the possibility of redistribution of the drug after death within and among different body compartments (see Ref 10). On the other hand, the autopsied brain approach offers the advantage of direct measurement of cocaine as well as its metabolites in discrete brain regions.

To our knowledge, no information is available with respect to the regional distribution of cocaine and its metabolites in brain of human users of the drug with the exception of a preliminary study of two autopsied cases in which only a small number of extra-striatal brain areas were examined (11). In the present investigation, we measured concentrations of cocaine and its metabolites benzoylecgonine, ecgonine methylester, norcocaine, and cocaethylene in 15 autopsied brain areas of 14 human chronic users of cocaine. Cases were selected from subjects who spanned the maximum range of brain drug concentrations and included subjects who died from drug overdose and those from other causes (e.g., trauma, hypertensive cardiac disease) in order to establish whether the cocaine regional distribution might be different in subjects who had recently taken a high, toxic dose of the drug as compared with those who probably did not use the drug immediately before death. We report that cocaine and its metabolites are relatively homogeneously distributed within the brain of chronic human users of the drug.

Subjects and Methods

Cocaine users—Postmortem brain material from a total of 14 users of cocaine was obtained from medical examiner offices in the

[1] Division of Forensic Toxicology, Office of the Armed Forces Medical Examiner, Division of Forensic Toxicology, Armed Forces Institute of Pathology, Washington, DC.
[2] Santa Clara County Medical Examiner-Coroner, San Jose, CA and Stanford University, Palo Alto, CA.
[3] Department of Pathology (Neuropathology), Sunnybrook Hospital, Toronto, Canada.
[4] Office of the Hillsborough County Medical Examiner, Tampa, FL.
[5] Office of the Medical Examiner of District 9, Orlando, FL.
[6] Department of Pathology, University of Maryland, Baltimore, MD.
[7] Human Neurochemical Pathology Laboratory, Centre for Addiction and Mental Health, Clarke Division, Toronto, Canada.
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U.S. and Canada. Drug history, brain neuropathological findings, and results of toxicological analyses of cocaine and its metabolites in blood and in one autopsied brain area (caudate nucleus) of 12 of the 14 cocaine users have been previously reported (12). Cases were included for study only if toxicological analysis indicated the presence of cocaine and/or metabolites in brain and in blood or urine in the absence of other drugs of abuse with the exception of alcohol. Information from the next of kin, informants, and from the case records indicated that all of the subjects, with the possible exception of case #570, had used cocaine for at least one year. A simple, standardized brain handling protocol was used at each medical examiner site. At autopsy, one half-brain was fixed in formalin fixative for neuropathological analysis, whereas the other entire half brain, as well as a sample of cardiac blood, was immediately frozen at −80°C until biochemical analysis. Scalp hair samples for drug analyses could be obtained from 7 of the 14 cocaine users. This study was approved by the University of Toronto institutional review committee.

Brain dissection for neurochemical analysis—Cerebral cortical subdivisions were excised using the Brodmann classification. Following dissection of the cerebral cortical subdivisions, approximately 2.5 mm-thick coronal sections of the brain were taken beginning with the anterior tip of the head of the caudate nucleus to the tail of the caudate. Subcortical brain areas were dissected using the Atlas of Riley (13) and as previously described (14). In areas containing significant amounts of both gray and white matter (e.g., cerebral cortex), the gray matter was carefully dissected out from the tissue for biochemical analysis. Because of the amount of tissue required for drug level analysis, measurements were not conducted in the small areas of the nucleus accumbens and the substantia nigra.

Measurement of concentrations of cocaine and its metabolites—Postmortem brain samples were dissected from the cocaine users and analyzed for cocaine and its metabolites (benzoylecgonine, ecgonine methylester, norcocaine, and cocaethylene). Approximately 100 mg of tissue was homogenized in 5 mL of 100 mM phosphate buffer at pH 6 (±0.5), using a Polytron homogenizer (Brinkman) at 10,000 rpm for 5 to 10 sec. Deuterated internal standards, for cocaine and each metabolite, were added prior to homogenization. Samples were allowed to sit at room temperature for 30 min, then centrifuged at 3000 × g for 5 min prior to drug extraction. Cocaine and its metabolites were extracted by solid phase extraction (Worldwide Monitoring, Clean Screen extraction columns #ZCDAU020, United Chemical Technologies, Bristol, PA) using the manufacturer's protocol. Briefly, the columns were conditioned by successive addition of 3 mL of methanol, 3 mL of deionized water and 3 mL 100 mM pH 6.0 phosphate buffer. Centrifuged samples were added to the column, allowed to drain by gravity and washed with successive additions of 3 mL deionized water, 3 mL 100 mM HCl and 2 mL methanol, after which the columns were dried under vacuum (>10 mm Hg) for 10 min. The analytes were eluted using 5 mL of a methylene chloride/isopropyl alcohol/ammonium hydroxide (78/20/2) solution and the eluent dried under nitrogen in 50°C water bath. The dried samples were derivatized using 50 μL pentafluoropropanol and 50 μL of ethyl acetate for analysis using gas chromatography/mass spectrometry (GC/MS). Sample quantitation was performed by GC/MS using a 5890 gas chromatograph (Hewlett-Packard (HP) Palo Alto, CA) interfaced to a 5989A mass spectrometer (HP). The GC was equipped with a 15 m, 0.25 mm i.d., 0.25 μm film-thickness, fused silica cross-linked methyl silicone, capillary column (DB-5MS-J&W Scientific, Folsom, CA) with helium (1 mL/min) as the carrier gas. Spitless injections were performed with the GC oven temperature programmed at 120°C (hold 1 min) to 260°C at 20°C/min (hold 4 min), injector temperature was 285°C. The mass spectrometer was operated in the selected ion monitoring (SIM) mode with the transfer line and source at 285°C and 200°C respectively. Samples were quantitated using a 6 point standard curve for each analyte. The limits of detection were 0.1 ng/mL for cocaine and its primary metabolites and 0.5 ng/mL for cocaethylene.

Results

Subject characteristics.—Table 1 shows the ages, postmortem intervals (interval between death and freezing of the half-brain at −80 °C), sex, estimate of the duration of drug use from the clinical

TABLE 1—*Patient information and drug histories for human cocaine users.*

Case	Age (yrs)	Autopsy Interval (h)	Sex	Estimated Duration (yrs)	Suspected/Known Cause of Death
234	26	18	M	1–2	cocaine intoxication (acute cocaine overdose)
344	31	16	M	6	cocaine intoxication
388	21	6	M	2–3	hypertensive cardiac disease with chronic cocaine use a contributing factor
533	36	26	M	8	cocaine intoxication
544	31	22	M	>2	cocaine intoxication
506	39	24	M	3	hypertensive cardiac disease with metoprolol/diltiazem intoxication
626	40	9	M	>10	gunshot wound to chest
629	36	24	M	>2	cocaine intoxication
648	30	20	F	>1	rupture aneurysm of right carotid artery with chronic cocaine use a contributing factor
505	26	18	F	8	cocaine intoxication
528	36	24	M	3	cocaine intoxication
573	40	16	M	>2	multiple stab wounds to chest
570	44	16	M	?	multiple injuries sustained in automobile accident
631	70	10	M	55	atherosclerotic cardiovascular disease

NOTE: Twelve of the 14 cocaine users were originally described in Wilson et al. (12).

case histories, and the suspected (acute cocaine intoxication) or known (e.g., trauma) cause of death. As shown in Table 1, cocaine intoxication (i.e., acute cocaine overdose) was the suspected cause of death in seven of the 14 cocaine users. The suspected or known causes of death of the remaining subjects were hypertensive cardiac disease with cocaine or metoprolol/diltiazem as a contributing factor ($n = 2$), ruptured carotid artery aneurysm with cocaine as a contributing factor ($n = 2$), atherosclerotic cardiovascular disease ($n = 1$), and trauma ($n = 2$). All subjects were believed to have died suddenly with the exception of case #344 who was brought to the hospital two days before death. Toxicological analysis for drugs of abuse other than cocaine and its metabolites in brain, blood, and hair revealed the presence of alcohol in blood of cases #505, 528, and 573, and 6-acetylmorphine (a heroin derivative) in hair of case #344. Cocaethylene (the transesterification condensation product of cocaine and ethanol) was observed in brain of cases #528, 544, 533, and 573 (see Table 2).

Blood and hair cocaine and metabolite concentrations—Table 2 shows the concentrations of cocaine and its metabolites in blood and hair of the 14 cocaine users. With the exception of two cases (#388 and #506), who tested positive for cocaine in urine, but not blood, each subject tested positive for cocaine and/or its major metabolite benzoylecgonine in blood, with a marked (approximately 1000-fold) range of drug concentrations among the individual subjects. The presence of cocaine and/or metabolites in blood or urine of all 14 subjects suggests that each individual probably had taken the drug within 48–72 h before death.

Hair samples (either single sample of tangle of hair or consecutive 0.5 in. segments) were taken for measurement of cocaine and metabolites from the 7 of the 14 cocaine users for which hair samples were available. As shown in Table 2, analysis for cocaine and benzoylecgonine indicated the presence of cocaine and benzoylecgonine in all samples examined with the exception of case #506, for which benzoylecgonine but not cocaine was detected. The presence of cocaine in consecutive hair segments from the scalp provides confirmation that these subjects had used the drug chronically.

Brain cocaine and metabolite concentrations.—As the brain concentrations of cocaine and its metabolites varied markedly among the individual patients, ranging over three orders of magnitude, the regional brain drug concentrations in 15 brain areas of each of the 14 cocaine users are shown in Table 3.

Brain concentrations of the parent compound, cocaine, showed the most marked intersubject variation, ranging from a high of approximately 190 nmol/g (tissue wet weight) in case #234 to low concentrations (less than 1 nmol/g) in case #344 which could be detected in only five of the 15 examined brain areas of this subject. The very high brain concentrations of cocaine in case #234 reflects the high blood concentrations of the drug in this individual (50 mg/L; see Table 2) who was known to have taken the drug within 4 h of death (Table 1). Case #344, who had very low concentrations of cocaine and its metabolites throughout the brain was known to have been abstinent from the drug for the 48 h prior to death. Less variation was observed for concentrations of benzoylecgonine among the different subjects (range 0.29 to 10.6 nmol/g), with concentrations of this cocaine metabolite in case #234 who had the highest brain cocaine level, not strikingly higher than concentrations of benzoylecgonine in the other 13 subjects. Concentrations of ecgonine methylester, norcocaine, and cocaethylene could be

TABLE 2—*Concentrations of cocaine and metabolites in blood and hair.*

Case		Cocaine	BZE	EME	Cocaethlylene	Norcocaine
234	Blood	50.0	75.0	ND	ND	ND
	Hair (NA)					
344	Blood	0.25	0.70	ND	ND	ND
	Hair (one)†	44.7	12.2			
388*	Blood	ND	ND	NE	NE	NE
	Hair (three)	117–159	6.0–8.1			
533	Blood	0.13	ND	NE	NE	NE
	Hair (tangle)	6.23	1.62			
544	Blood	5.40	6.20	NE	NE	NE
	Hair (NA)					
506*	Blood	ND	ND	NE	NE	NE
	Hair (tangle)	ND	8.25			
626	Blood	0.03	3.55	0.74	ND	ND
	Hair (four)	36.8–69.6	0.5–3.2			
629	Blood	0.05	ND	NE	NE	NE
	Hair (NA)					
648	Blood	0.19	4.79	0.91	ND	ND
	Hair (seven)	102–183	17.2–50.4			
505	Blood	0.08	ND	NE	NE	NE
	Hair (NA)					
528	Blood	5.42	2.65	2.33	0.04	0.15
	Hair (NA)					
573	Blood	0.01	0.06	0.02	0.01	ND
	Hair (four)	5.5–24.2	0.7–1.8			
570	Blood	0.06	0.02	NE	NE	NE
	Hair (NA)					
631	Blood	0.32	5.32	NE	NE	NE
	Hair (NA)					

NOTE: Values in blood and hair are mg/L and ng/mg, respectively. * Cocaine not detected in blood, but present in urine; † Number of half inch segments of hair analyzed; BZE (benzoylecgonine), EME (ecgonine methylester, NA, not available; ND, not detected; NE, not examined. The blood alcohol concentrations in both subjects #528 and #573 were 200 mg/L. Twelve of the 14 cocaine users were originally described in Wilson et al. (12).

TABLE 3—*Regional distribution of molar concentrations of cocaine and its metabolites benzoylecgonine, ecgonine, methylester, norcocaine, and cocaethylene in autopsied brain of 14 human cocaine users.*

	234	528	626	631	544	648	388	505	533	570	629	506	573	344
Cocaine:														
Front	136	66.5	1.36	3.88	53.0	1.20	0.69	0.83	1.63	0.69	0.43	0.32	0.58	0.71
Temp	159	85.4	1.37	4.17	82.4	1.21	0.73	0.71	1.60	0.79	0.44	0.34	0.34	0.48
Occ	190	62.3	1.82	5.58	70.4	1.31	---	0.75	3.35	0.91	0.41	0.31	0.27	0.22
Par	165	74.0	1.16	4.55	63.4	1.28	0.69	0.59	1.94	0.82	0.41	0.32	0.33	0.11
Cing Ctx	166	57.2	1.54	3.10	64.7	0.99	0.62	1.01	1.78	0.63	0.41	0.32	0.22	0
Cereb	148	53.9	0.98	3.94	58.9	0.86	0.59	0.66	1.69	0.62	0.29	0.29	0.16	0.01
Caudate	118	93.9	0.84	5.67	68.6	0.86	0.86	0.73	1.90	---	0.62	0.41	0.23	0
Putamen	187	113	1.73	6.65	85.6	1.28	0.97	1.13	2.64	1.25	0.58	0.24	0.19	0
Parolf	87.8	47.3	1.53	8.53	47.4	0.96	0.60	0.78	2.04	0.57	0.49	0.30	0.19	0
GPI	148	86.4	1.56	5.90	63.0	1.04	0.72	0.81	2.93	0.82	0.47	0.40	0.15	0
GPE	137	92.8	1.37	8.42	67.1	1.08	0.79	0.86	3.16	0.89	0.48	0.32	0.16	0
Hippo	143	81.8	1.05	6.06	69.9	0.50	0.68	0.47	2.07	0.59	0.53	0.33	0.14	0
Hypo	100	73.0	0.78	3.31	48.6	0.64	0.62	0.49	1.57	0.44	0.40	0.29	0.17	0
MDTH	196	132	1.41	8.13	74.0	1.04	0.74	0.62	2.08	0.82	0.44	0.38	0.14	0
NPM	161	132	1.48	9.88	66.9	0.85	0.68	0.47	1.30	0.63	0.43	0.35	0.14	0
Benzoylecgonine														
Front	4.78	6.01	3.60	5.04	1.39	5.22	8.28	2.34	2.39	0.33	0.40	0.97	0.35	1.10
Temp	6.01	8.09	2.82	4.94	2.04	5.62	9.19	2.13	2.11	0.28	0.25	0.95	0.30	1.15
Occ	6.43	9.48	4.55	5.93	2.53	5.86	---	2.26	2.41	0.44	0.26	0.98	0.39	1.10
Par	5.55	9.20	3.22	4.90	1.73	5.85	8.90	2.49	2.74	0.32	0.21	0.96	0.35	1.02
Cing Ctx	6.33	8.63	3.95	4.16	1.94	6.02	9.98	2.44	2.36	0.31	0.31	1.01	0.34	1.16
Cereb	6.89	8.80	4.12	5.23	3.27	6.05	9.76	2.56	2.33	0.31	0.40	0.99	0.54	0.99
Caudate	5.81	9.14	4.25	4.62	2.24	5.32	10.6	2.49	2.46	---	0.27	1.02	0.33	1.23
Putamen	6.41	9.15	4.43	5.71	2.60	5.63	8.04	2.86	2.37	0.29	0.22	0.95	0.34	1.40
Parolf	3.44	6.85	1.92	3.95	0.86	4.08	6.58	2.33	0.68	0.23	0.18	0.73	0.33	1.20
GPI	4.64	8.47	3.24	3.37	1.51	5.11	7.61	2.89	1.54	0.29	0.20	0.86	0.33	1.39
GPE	5.09	8.66	3.48	4.38	2.07	5.04	8.75	2.66	1.70	0.33	0.20	0.90	0.35	1.32
Hippo	5.38	8.43	3.61	4.36	2.17	6.01	9.61	2.23	1.69	---	0.34	1.00	0.30	1.10
Hypo	5.16	8.58	3.73	4.85	1.82	6.50	9.53	2.47	1.93	0.31	0.26	0.97	0.30	1.11
MDTH	7.14	9.88	4.66	6.41	3.05	5.31	8.91	2.57	2.49	0.30	0.24	0.96	0.33	1.21
NPM	7.22	9.82	5.07	4.74	2.67	5.81	9.93	2.38	2.42	0.32	0.30	0.98	0.30	1.10
Ecgonine methylester														
Front	5.95	7.41	3.92	4.50	4.83	2.02	5.62	0.59	2.41	0.17	0.22	0.74	0.30	0.52
Temp	7.77	8.96	3.66	5.17	7.61	2.31	7.02	0.65	2.14	0.19	0.26	0.98	0.24	0.55
Occ	---	9.42	4.61	5.52	9.18	2.13	---	0.69	2.49	0.21	0.24	0.87	0	0.53
Par	6.91	9.00	3.09	4.58	8.97	2.06	6.23	0.62	2.21	0.20	0.21	0.76	0	0.47
Cing Ctx	8.56	8.72	---	3.70	6.85	2.18	5.66	0.76	2.17	0.23	0.28	0.80	0	0.48
Cereb	13.4	12.2	4.09	7.27	13.3	2.32	5.97	0.67	2.60	0.27	0.38	0.73	0	0.47
Caudate	11.2	11.2	3.94	5.15	10.5	2.04	5.18	0.83	2.36	---	0.33	0.74	0	0.58
Putamen	12.0	13.0	5.30	6.54	11.7	2.33	6.35	0.74	2.77	0.22	0.32	0.91	0	0.49
Parolf	6.16	6.09	3.13	5.05	6.63	1.65	4.39	0.66	1.67	0.19	0.24	0.49	0	0.49
GPI	8.06	9.94	4.40	---	8.74	1.91	5.13	0.72	2.00	0.23	0.29	0.71	0	0.51
GPE	10.2	11.4	4.87	5.37	11.0	1.94	5.34	0.87	2.19	0.25	0.27	0.78	0	0.53
Hippo	8.15	8.54	3.86	4.96	8.11	1.41	4.76	0.77	2.27	0.21	0.27	0.76	0	0.51
Hypo	12.9	8.30	3.00	3.41	6.82	1.73	3.93	0.61	1.87	0.19	0.33	0.53	0	0.46
MDTH	14.1	11.9	5.02	6.29	11.2	2.03	7.09	0.72	2.35	0.24	0.26	0.82	0	0.56
NPM	9.92	11.4	8.14	4.50	9.07	1.94	4.93	0.70	2.09	0.24	0.27	0.69	0	0.51

TABLE 3—*Continued*

	234	528	626	631	544	648	388	505	533	570	629	506	573	344
Norcocaine														
Front	7.70	2.83	0.19	0.17	0.74	0.19	0	0.15	0.28	0	0	0	0	0
Temp	6.98	1.88	0.16	0.16	1.78	0.27	0	0.16	0.34	0	0	0	0	0
Occ	6.91	1.53	0.16	0.22	0.95	0.18	---	0.15	0.44	0	0	0	0	0
Par	6.82	1.77	0.12	0.16	1.16	0.19	0	0.14	0.24	0	0	0	0	0
Cing Ctx	8.75	1.92	0.17	0.16	1.33	0.15	0	0.19	0.45	0	0	0	0	0
Cereb	7.06	1.32	0.11	0.21	1.00	0.11	0	0.14	0.27	0	0	0	0	0
Caudate	4.65	1.36	0.18	0.21	0.84	0.06	0	0.21	0.26	---	0	0	0	0
Putamen	10.1	2.05	0.22	0.28	1.19	0.08	0	0.17	0.36	0	0	0	0	0
Parolf	3.21	1.58	0.25	0.61	1.12	0.13	0	0.16	0.39	0	0	0	0	0
GPI	5.72	1.66	0.21	0.21	1.36	0.10	0	0.15	0.27	0	0	0	0	0
GPE	6.40	1.27	0.25	0.32	1.01	0.18	0	0.22	0.33	0	0	0	0	0
Hippo	10.1	1.27	0.22	0.17	1.13	0.06	0	0.18	0.26	0	0	0	0	0
Hypo	4.15	1.16	0.13	0.23	1.31	0.11	0	0.18	0.28	0	0	0	0	0
MDTH	8.89	1.52	0.22	0.20	1.84	0.16	0	0.18	0.51	0	0	0	0	0
NPM	8.35	1.68	0.24	0.40	1.06	0.06	0	0.18	0.26	0	0	0	0	0
Cocaethylene														
Front	0	0.46	0	0	0.61	0	0	0	0.13	0	0	0	0.24	0
Temp	0	0.62	0	0	1.08	0	0	0	0.12	0	0	0	0.25	0
Occ	0	0.38	0	0	0.94	0	---	0	0.29	0	0	0	0.25	0
Par	0	0.47	0	0	0.78	0	0	0	0.14	0	0	0	0.25	0
Cing Ctx	0	0.37	0	0	0.85	0	0	0	0.19	0	0	0	0.25	0
Cereb	0	0.33	0	0	0.73	0	0	0	0.13	0	0	0	0.21	0
Caudate	0	0.63	0	0	0.80	0	0	0	0.14	---	0	0	0.26	0
Putamen	0	0.58	0	0	0.95	0	0	0	0.13	0	0	0	0.29	0
Parolf	0	0.30	0	0	0.80	0	0	0	0.16	0	0	0	0.28	0
GPI	0	0.68	0	0	0.87	0	0	0	0.16	0	0	0	0.29	0
GPE	0	0.60	0	0	0.88	0	0	0	0.20	0	0	0	0.29	0
Hippo	0	0.52	0	0	0.84	0	0	0	0.10	0	0	0	0.26	0
Hypo	0	0.46	0	0	0.63	0	0	0	0.11	0	0	0	0.25	0
MDTH	0	0.82	0	0	0.99	0	0	0	0.15	0	0	0	0.31	0
NPM	0	0.82	0	0	0.97	0	0	0	0.12	0	0	0	0.25	0
Cocaine + metabolites														
Front	155	83.2	9.07	13.6	60.6	8.63	14.6	3.90	6.85	1.19	1.05	2.03	1.47	2.33
Temp	179	105	8.01	14.4	94.9	9.40	16.9	3.64	6.32	1.26	0.95	2.27	1.12	2.18
Occ	203	83.1	11.1	17.2	84.0	9.48	---	3.85	8.97	1.57	0.90	2.16	0.91	1.85
Par	185	94.4	7.60	14.2	76.1	9.39	15.8	3.83	7.27	1.34	0.83	2.04	0.94	1.60
Cing Ctx	190	76.9	5.66	11.1	75.7	9.34	16.3	4.40	6.95	1.16	1.00	2.12	0.82	1.65
Cereb	175	76.5	9.31	16.6	77.3	9.35	16.3	4.02	7.02	1.20	1.07	2.01	0.90	1.46
Caudate	140	116	9.20	15.6	83.1	8.28	16.6	4.26	7.12	---	1.21	2.17	0.82	1.80
Putamen	216	138	11.7	19.2	102	9.31	15.4	4.90	8.27	1.76	1.12	2.10	0.82	1.89
Parolf	101	62.1	6.83	18.1	56.8	6.82	11.6	3.93	4.95	0.99	0.91	1.52	0.80	1.69
GPI	166	107	9.43	9.47	75.5	8.16	13.5	4.57	6.89	1.33	0.96	1.96	0.77	1.90
GPE	158	115	9.98	18.5	82.0	8.24	14.9	4.61	7.57	1.48	0.95	2.00	0.80	1.85
Hippo	167	101	8.74	15.5	82.1	7.98	15.0	3.65	6.40	0.80	1.14	2.09	0.70	1.62
Hypo	123	91.5	7.63	11.8	59.2	8.98	14.1	3.74	5.76	0.94	0.99	1.79	0.72	1.58
MDTH	226	156	11.3	21.0	91.1	8.55	16.7	4.10	7.58	1.36	0.94	2.16	0.79	1.77
NPM	186	156	14.9	19.5	80.7	8.66	15.5	3.72	6.19	1.19	1.00	2.02	0.69	1.62

NOTE: Values are nmol/g tissue wet weight. Abbreviations: front (frontal cortex, Brodmann area 10), temp (temporal cortex, Brodmann area 21), occ (occipital cortex, Brodmann area 17), par (parietal cortex, Brodmann area 7b), cing ctx (cingulate cortex, Brodmann area 24), Cereb (cerebellar cortex), parolf (parolfactory cortex, Brodmann area 25), GPI (internal globus pallidus), GPE (external globus pallidus), hippo (Ammon's horn of hippocampus), hypo (hypothalamus), MDTH (medial-dorsal thalamus), NPM (medial pulvinar thalamus)—not examined.

detected in most brain areas of 13, 8, and 4 of the 14 cocaine users, respectively. In terms of the relative concentrations of cocaine and its metabolites in brain of the individual subjects, cocaine was the highest concentration in 6 of the 14 subjects, whereas concentrations of benzoylecgonine and ecgonine methylester were the highest in six and two subjects, respectively.

Examination of the regional distribution of cocaine and its metabolites did not disclose any marked regional differences among the 15 brain areas in any of the 14 cocaine users (Table 3). This included subjects for which the suspected cause of death was cocaine intoxication and who were believed to have died shortly after taking the drug (cases #234, 528, 544, 505, 533, and 629) and those three subjects who had the highest brain cocaine concentrations (#234, 528, and 544). In the 14 subjects, the brain regions having the highest cocaine concentrations were the following: cerebral cortex ($n = 5$), putamen ($n = 4$), thalamus ($n = 3$), and caudate ($n = 2$).

In order to compare the regional distribution differences of cocaine and its metabolites across the 14 subjects, who spanned a wide range of concentrations, the level of the drug in each region of individual subjects was normalized to the mean drug level of all 15 regions examined, which was assigned a value of 100. As can be seen in Table 4, the magnitude of the differences in normalized values among the 15 brain regions was only slight, ranging from mean concentrations 27% below (cocaine, hypothalamus) and 25% above (cocaine, putamen) the mean 100 level for cocaine, benzoylecgonine, and ecgonine methylester. Somewhat less variation was observed for the regional distributions of the sum of normalized "total cocaine," i.e., the sum of cocaine and its examined metabolites, in the 14 subjects (range of means: 81 to 117; see Table 4 and Fig. 1). Total cocaine concentrations were also determined in the subgroup of cocaine users who had most likely died very shortly after cocaine overdose (for which postmortem brain drug concentrations would best approximate those in living brain). As with the total group of 14 subjects, little variation was observed for the regional distribution of the sum of normalized total cocaine for the entire subgroup of six subjects (#234, 528, 544, 505, 533, and 629) suspected of dying from cocaine intoxication who had died suddenly (range of means: 74 [parolfactory cortex] to 123 [putamen]) or for the three subjects (#234, 528, 544) with the highest cocaine concentrations (range of means: 75 [parolfactory cortex] to 132 [medial dorsal thalamus]) (see Fig. 1).

Discussion

The major finding of our investigation is that concentrations of cocaine and of its major metabolites show little regional heterogeneity in postmortem brain of chronic users of cocaine.

Cocaine metabolizes in the body to benzoylecgonine, ecgonine methylester, and norcocaine (15). In cases in which cocaine is used

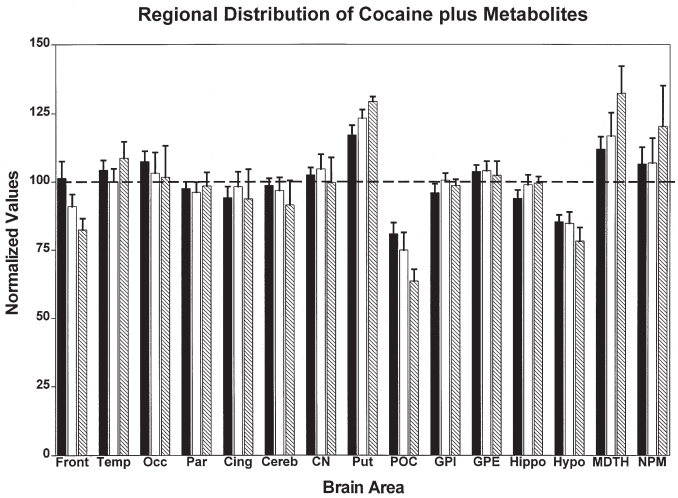

FIG. 1—*The drug concentrations shown in Table 3 were used to calculate a mean drug level of all 15 brain regions examined in each subject which was assigned a value of 100. Values represent the mean ± S.E. of the normalized mean concentrations of cocaine plus all examined metabolites (benzoylecgonine, ecgonine methyl ester, norcocaine, and cocaethylene) in 15 brain regions of the total group of 14 human cocaine users (solid black bar), six cocaine users (#234, 528, 544, 505, 533, and 629) who likely died suddenly from cocaine intoxication (open bar) and the three cocaine users (#234, 528, and 544) who had the highest brain cocaine concentrations (striped bar)(see Results section for details). Abbreviations: front (frontal cortex, Brodmann area 10), temp (temporal cortex, Brodmann area 21), occ (occipital cortex, Brodmann area 17), par (parietal cortex, Brodmann area 7b), cing ctx (cingulate cortex, Brodmann area 24), Cereb (cerebellar cortex), CN (caudate), Put (putamen), POC (parolfactory cortex, Brodmann area 25), GPI (internal globus pallidus), GPE (external globus pallidus), hippo (Ammon's horn of hippocampus), hypo (hypothalamus), MDTH (medial-dorsal thalamus), NPM (medial pulvinar thalamus).*

TABLE 4—*Regional distribution of normalized values of cocaine, its metabolites, and the sum of cocaine and its metabolites (cocaine + metabolites) in 14 human cocaine users.*

Brain Region	Cocaine	Benzoylecgonine	EME	Norcocaine	Cocaethylene	Cocaine + metabolites
Frontal Cortex	117 ± 17	97 ± 5	88 ± 5	103 ± 13	84 ± 4	101 ± 6
Temporal Cortex	111 ± 8	95 ± 2	98 ± 4	113 ± 15	104 ± 11	104 ± 4
Occipital Cortex	111 ± 6	111 ± 4	103 ± 2	100 ± 8	117 ± 27	107 ± 4
Parietal Cortex	96 ± 6	98 ± 3	91 ± 3	91 ± 9	93 ± 2	98 ± 3
Cingulate Cortex	87 ± 9	103 ± 2	96 ± 3	108 ± 8	97 ± 11	94 ± 4
Cerebellar Cortex	74 ± 6	116 ± 6	118 ± 5	82 ± 4	78 ± 5	99 ± 3
Caudate	101 ± 6	105 ± 2	107 ± 3	82 ± 8	101 ± 5	102 ± 3
Putamen	125 ± 7	106 ± 3	118 ± 3	108 ± 9	104 ± 6	117 ± 4
Parolfactory Cortex	92 ± 7	71 ± 5	79 ± 3	116 ± 21	91 ± 12	81 ± 4
Interior Globus Pallidus	106 ± 5	89 ± 4	97 ± 2	92 ± 5	110 ± 6	96 ± 3
Exterior Globus Pallidus	110 ± 6	95 ± 3	107 ± 3	110 ± 9	114 ± 6	104 ± 2
Hippocampus	88 ± 6	98 ± 3	93 ± 3	91 ± 11	91 ± 8	94 ± 3
Hypothalamus	73 ± 3	98 ± 2	87 ± 6	84 ± 6	83 ± 6	85 ± 3
Medial Dorsal Thalamus	109 ± 7	110 ± 4	115 ± 4	119 ± 9	122 ± 11	112 ± 5
Pulvinar Medial Thalamus	100 ± 9	109 ± 4	105 ± 7	103 ± 12	111 ± 16	106 ± 6

NOTE: The drug concentrations shown in Table 3 were used to calculate a mean drug level of all 15 brain regions examined in each subject which was assigned a value of 100. Values represent the mean ± S.E. of the normalized drug concentrations in the 14 human cocaine users.

in combination with ethanol, the transesterification product of cocaine and ethanol, cocaethylene, is also formed as a metabolite (16). Cocaine readily crosses the blood brain barrier with animal studies showing that brain concentrations of cocaine are about four times those of plasma at peak plasma concentrations (17). Benzoylecgonine, however, is restricted in transport across the blood brain barrier, found in brain concentrations approximately one tenth those of plasma at peak plasma concentration, and, when detected in brain, appears to be derived from the metabolism of cocaine that was transported into the brain (18).

Although postmortem blood concentrations of cocaine and its metabolites cannot be used to predict accurately the interval between the last drug administration and death (see Ref 10), Spielhler and Reed (11) have obtained autopsy data suggesting that the brain/blood ratio of cocaine can provide a very general estimate of this time inverval, with subjects having high brain/blood cocaine ratios (median 3.8; range, 0.65 to 155) likely to have used the drug recently (within hours before death). The brain/blood ratios for cocaine for the subjects examined in our investigation fall within this general range with the exception of cases #388 and #506 who tested positive for cocaine in brain, but not blood. This likely reflected, in these subjects, a long interval (days) between the last cocaine administration and death.

A limitation of our study is the possibility that the regional pattern of concentrations of cocaine and its metabolites as determined in autopsied brain might not reflect the distributional profile in living brain. Thus, during the interval between death and freezing of the brain, there might have been some redistribution of the drug among the different brain areas and between the brain, blood, and CSF components that could have minimized, to some extent, a preferential accumulation of cocaine occurring in the striatum at an earlier point in time. Some redistribution of cocaine and its metabolites among different brain regions also probably occurred premortem (i.e., during the time interval between the last dose of the drug and death), and it is quite possible that an initial local increase in brain concentration of cocaine in, for example, the striatum, was lost over time. However, it can be argued that during the time following drug administration, the dopamine-rich brain areas should still have retained some preferential residual accumulation which could be detected postmortem if behaviorally relevant doses of cocaine are, in fact, principally accumulated and retained in the striatum.

A second limitation of our investigation is that absolute documentation of the interval between the last dose of cocaine and death could be obtained for only one case (#234), in which the drug taking was witnessed four hours preceding death. However, it is likely that the interval between last drug taking and death was also short for the other five subjects suspected of having a sudden death from cocaine intoxication (528, 544, 505, 533, and 629) including, especially, case #505, who was found with a crack pipe under his body (12).

Comparison of cocaine concentrations in dopamine/dopamine transporter-rich vs. -poor brain areas in the individual cocaine users revealed that drug concentrations in the putamen were higher than those in many of the other brain areas examined (Table 3). However, there were other dopamine-poor brain areas having similarly "high" drug concentrations. For example, in case #234, who had the highest brain cocaine concentration, the level of cocaine in the putamen (187 nmol/g) was similar to that in the anatomically distant dopamine transporter-poor occipital cortex (190 nmol/g) and medial dorsal thalamus (196 nmol/g) (Table 3). Although, as shown in Fig. 1, mean normalized "total cocaine" concentrations were, on average, higher in the putamen than in other regions in the total cocaine user group ($n = 14$) and, in the sudden death cocaine intoxication subgroup ($n = 6$), the average magnitude of the increase above mean drug level was only modest (approximately 15 to 25%) and was similar to that observed for the dopamine transporter-poor medial dorsal thalamus (see Fig. 1). Most importantly, concentrations of cocaine and its metabolites in the cocaine users were clearly not high in the caudate nucleus, a striatal subdivision, like the putamen, enriched in the dopamine transporter, with drug concentrations in caudate nucleus and in the anatomically distant, dopamine transporter-poor cerebellar cortex being similar in these cases (see Tables 3 and 4, Fig. 1). Surprisingly, concentrations of cocaine and its metabolites were, on average, slightly low in the parolfactory cortex (Brodmann area 25), the only area of the human cerebral cortex that contains appreciable concentrations of dopamine (parolfactory cortex, Brodmann area 25; Ref 19) (Fig. 1).

In principle, the relatively homogeneous regional distribution pattern of cocaine and metabolites observed in postmortem brain of the drug users could be explained by an initial selective binding of cocaine to the striatal dopamine transporter *in vivo* followed by a rapid (e.g., minutes) dissociation and redistribution to other brain areas or by an initial non-selective distribution of cocaine to, and retention in all brain areas, with the later occurring because the striatal dopamine transporter is saturated at high drug concentrations. Although our postmortem study cannot distinguish between these two possibilities, we suggest that the latter possibility is not unreasonable. In fact, this speculation is supported by PET imaging data in the baboon (8) in which a homogeneous brain regional distribution of ^{11}C-cocaine is observed immediately following a high "pharmacological" dose (8 mg) of the drug. Our data are also consistent with the results of, to our knowledge, the only other postmortem brain study of cocaine users conducted to date (11), in which concentrations of cocaine and benzoylecgonine in a dopamine transporter-rich area (substantia nigra) were similar to those in dopamine transporter-poor brain regions (frontal, occipital, cerebellar cortices, medulla, and spinal cord) in two subjects.

There continues to be an assumption in the scientific community that cocaine, which binds to the dopamine transporter, must therefore accumulate and be retained selectively in dopamine transporter-rich brain areas in human users of the drug. This assumption is probably based in part on the striking single photon emission computed tomography (SPECT) demonstration that the radioiodinated high affinity cocaine analogue [^{123}I] β-CIT is selectively retained in human striatum at least 32 h following administration (20). While animal data indicate that this regional selectivity is certainly true for low tracer doses of cocaine, we question this assumption for much higher behaviorally relevant doses of the low affinity parent compound used in the human and, especially, in those users, as in our investigation, who take the drug on a chronic basis. Although, as mentioned above, it is not possible in any postmortem study to establish absolutely a regional brain drug profile occurring in living brain, our data suggest that cocaine will be distributed widely in brain of human chronic users of the drug. As such, consideration should be given to the potential pharmacological and neurotoxicological actions of cocaine in both striatal and extra-striatal brain regions.

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Additional information and reprint requests:
Stephen J. Kish, Ph.D.
Human Neurochemical Pathology Laboratory
Centre for Addiction and Mental Health
250 College Street
Toronto, Ontario
Canada M5T 1R8
Phone: 416-979-4747, ext. 2277
Fax: 416-979-6871
e-mail: Stephen_Kish@CAMH.net