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XENOBIOTIC-METABOLIZING ENZYMES IN HUMAN RESPIRATORY NASAL MUCOSA

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Abstract—Study of oxidative and non-oxidative xenobiotic-metabolizing enzymes was undertaken in microsomal and cytosolic fractions of two human livers, 10 individual and several pooled samples of human respiratory nasal mucosa obtained by surgical operation of male and female patients affected by hypertrophy of the inferior turbinates. The purity of nasal microsomes was checked by electron microscopy and marker enzyme assay. The pooled samples of respiratory nasal epithelium contained, relative to liver, a low amount of cytochrome P450 (about 25 pmol/mg protein) and associated biotransformation activities, and a low level of other components of the mixed-function oxidase system such as cytochrome b_5, NADH and NADPH-cytochrome *c* reductase however the NADH–cytochrome b_5 reductase activity was comparable to that of liver. The P450-dependent monooxygenase activities such as ethoxycoumarin *O*-deethylase, ethoxyresorufin *O*-deethylase and the dimethylnitrosamine *N*-demethylase were found in nearly all nasal microsomal specimens. The aniline hydroxylase and the aminopyrine or hexamethylphosphoramide *N*-demethylases were detected only in the pooled nasal samples. With regard to the non-oxidative enzymes, the activities of glutathione *S*-transferase, DT-diaphorase, epoxide hydrolase, UDP-glucuronyl-transferase, carbonyl reductase, benzaldehyde and propionaldehyde dehydrogenases, were investigated both in the individual and pooled nasal tissues and livers. These activities were similar in nasal and liver tissue, except for UDP-glucuronyltransferase which was not detected in nasal mucosa. The present findings demonstrate that the respiratory section of human nose contains a wide array of oxidative and non-oxidative enzymes, which could play a crucial role in the bioactivation or detoxication *in situ* of inhaled xenobiotics.

Nasal cavities represent a major port of entry into the body for volatile xenobiotics, including carcinogens, present in the environment. Many authors have demonstrated that respiratory and olfactory nose membranes of a variety of mammalian species contain P450† and are able to metabolize many chemical compounds [1–8].

The olfactory epithelium in monkey, dog, rabbit and rat shows a higher P450 content and higher microsomal monooxygenase activities than those of respiratory epithelium and, in many cases, even than liver [1–2].

The reason for the presence in the nose of this high drug-metabolizing activity, being second only to that of liver, is not clearly understood. Probably this high activity is necessary to rapidly remove the inhaled airborne odorants and toxicants thereby to maintain odour sensitivity [2]. In specific cases, however, a bioactivation *in situ* of inhaled promutagens and procarcinogens, could be a harmful consequence of this nasal biotransformation process of xenobiotics. Therefore, it is important to know in detail the catalytic activities and the substrate specificity of drug-metabolizing enzymes present in the nasal respiratory tract not only in animals such as rodents and monkeys but, if possible, in man.

Whereas in monkey respiratory membranes no measurable concentrations of P450 have been found [1], we have recently reported preliminary results showing that human respiratory epithelium contains detectable levels of P450 and associated-monooxygenase activities towards some substrates [9] and particularly towards the carcinogenic diethylnitrosamine [10].

The purpose of the present study was to extensively characterize the metabolic capacity of respiratory tissue using several specimens obtained from human patients affected by hypertrophy of inferior turbinates. Both oxidative and non-oxidative nasal enzyme activities were investigated and compared with the respective human hepatic levels and the corresponding data published for nasal mucosa of other animals.

MATERIALS AND METHODS

Chemicals. DEN, dichlorophenolindophenol, hexamethylphosphoramide, resorufin, 1-naphthol, acrylamide and rutin, were obtained from Fluka (Buchs,

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† Abbreviations used: P450, cytochrome P450; DEN, diethylnitrosamine; DENd, diethylnitrosamine *N*-deethylase; DMN, dimethylnitrosamine; DMNd, dimethylnitrosamine *N*-demethylase; AP, aminopyrine; APD, aminopyrine *N*-demethylase; ECOD, ethoxycoumarin *O*-deethylase; EROD, ethoxyresorufin *O*-deethylase; PROD, pentoxyresorufin *O*-depenthylase; pNPH, *p*-nitrophenol hydroxylase; HMPA, hexamethylphosphoramide; HMPAd, hexamethylphosphoramide *N*-demethylase; AnH, aniline hydroxylase; SAFO, safrole oxide; CDNB, 1-chloro-2,4-dinitrobenzene; UDP-GT, UDP-glucuronyltransferase; DCPIP, dichlorophenolindophenol; GST, glutathione *S*-transferase; EH, epoxide hydrolase; BeD, benzaldehyde dehydrogenase; PrD, propionaldehyde dehydrogenase; POOL, 10 pooled human respiratory nasal mucosa samples.

Table 1. Medical and smoking history of human subjects

Patient No.	Tissue	Age	Sex	Smoking habits	Drug treatments
16	Nasal	30	Male	Smoker	NA
17	Nasal	37	Male	Non-smoker	NA
18	Nasal	50	Male	Smoker	Vasoconstrictor drugs
19	Nasal	32	Male	NA	Vasoconstrictor drugs
20	Nasal	28	Male	Non-smoker	No drugs
21	Nasal	16	Female	Non-smoker	Anti-histaminic drugs
22	Nasal	46	Female	3 cigarettes/day	Anti-histaminic drugs
23	Nasal	19	Female	Non-smoker	NA
24	Nasal	38	Female	20 cigarettes/day	No drugs
25	Nasal	43	Female	Non-smoker	Decongestion drugs
HL6	Hepatic	23	Male	Non-smoker	No drugs
HL7	Hepatic	33	Male	Non-smoker	NA

NA, information not available.

Switzerland); dicumarol, DMN and ethoxycoumarin were purchased from EGA-Chemie (Steinheim, F.R.G.). 1-Chloro-2,4-dinitrobenzene was obtained from Eastman Kodak (Rochester, NY, U.S.A.); NADH and NADPH were purchased from Boehringer (Mannheim, F.R.G.). Ethoxyresorufin and pentoxyresorufin were synthesized from resorufin by ethylation with ethyl iodide and by pentylation with pentyl iodide, respectively [11]. SAFO was synthesized as described by Watabe and Akamatsu [12]. DEN was distilled before use. All other chemicals and solvents were of analytical grade and were obtained from common commercial sources.

Human liver and nasal tissue. Wedge biopsies of two human livers were obtained from patients undergoing cholecystectomy. The surplus of the material required for histological analysis was made available for our studies. Tissue (about 0.5 g) was promptly frozen and stored at −80°.

Nasal respiratory membranes were obtained after surgical removal for therapeutic purposes from 20 male and female patients affected by hypertrophy of the inferior turbinates. The experiments with nitrosamines were performed in a specially equipped area for handling mutagenic and carcinogenic compounds. The nitrosamine residues were degraded by adding aluminium–nickel alloy powder [13].

Nasal tissues were removed from patients and frozen in liquid N_2 within 10–15 min after removal and stored at −80°. Upon histopathological examination, no sign of leucocyte infiltration was noted in the samples investigated. The limited information available of the patients is presented in Table 1.

Preparation of microsomes. Liver and nasal mucosa microsomal fractions were prepared using an Ultra-Turrax homogenizer as previously described [5]. Nasal microsomes and 100,000 *g* supernatant fractions were prepared individually from 10 nasal specimens and collectively from the other 10 nasal samples (POOL) which were pooled together due to the low amount of available tissue (<0.5 g). Washed microsomes were resuspended in 20% glycerol/potassium phosphate buffer (50 mM, pH 7.4), 1 mM EDTA and stored at −80° until assayed. Cytosolic and microsomal protein concentrations were measured by the method of Lowry *et al.* [14] using bovine serum albumin as standard.

For electron microscopy, the resuspended microsomes in the above mentioned buffer were spun down at 100,000 *g* in an ultracentrifuge for 30 min and the supernatant removed. The pellets were rinsed once with buffer and fixed in 3% glutaraldehyde. After washing, the fragments were postfixed in 1% osmium tetroxide, dehydrated with ethanol and propylene oxide, and embedded in Epon. The sections were stained with uranyl acetate and lead citrate and examined in a Siemens Elmiskop 101 electron microscope.

Enzymic assays. Cytochrome P450 and cytochrome b_5 were determined by the method of Matsubara *et al.* [15]. NADH and NADPH–cytochrome *c* reductase were assayed by monitoring cytochrome *c* reduction at 550 nm [16]. NADH–cytochrome b_5 reductase was measured as described by Mihara and Sato [17] using potassium ferricyanide as electron acceptor. Cytochrome oxidase activity was measured by the method of Wharton and Tzagoloff [18]. DT-diaphorase and carbonyl reductase activities were assayed as earlier described [19, 20] following, at 600 nm, the reduction of DCPIP (40 μM) (as electron acceptor) inhibitable by 1 μM dicoumarol or 10 μM rutin, respectively. Microsomal EH activity was measured using the method of Watabe and Akamatsu [12] with SAFO (2 mM) as substrate. GST activity was determined spectrophotometrically using CDNB (1 mM) as substrate [21]. UDP-GT activity was quantitated according to MacKenzie and Hanninen [22] using 1-naphtol (50 μM) as substrate which is specific for a single form of UDP-GT (UDP-GT_1). Benzaldehyde and propionaldehyde dehydrogenase activities were assayed as described by Lindahl and Feinstein [23]. ECOD and pNPH activities were determined according to Aitio [24] and Reinke and Moyer [25], respectively. The concentration of ethoxycoumarin was 0.4 mM and that of *p*-nitrophenol was 0.1 mM. The extent of DEN deethylation was determined by HPLC as previously reported in detail at low (1 mM) and high (50 mM) DEN concentration [26]. AnH activity was measured according to Ko *et al.* [27]. The aniline concentration

Table 2. Characteristics of the microsomes of human respiratory nasal mucosa POOL

Parameters	Amount or specific activity	Recovery (% of homogenate)
Yield of protein*	2.84	4
NADPH–cytochrome *c* reductase†	32.6	35
Cytochrome *c* oxidase‡	99.8	12

The results are from microsomal and homogenate POOL of nasal samples.
* Values in mg/g nasal tissue.
† Values in nmol cytochrome *c* reduced/min × g tissue.
‡ Values in nmol cytochrome *c* oxidized/min × g tissue.

was 2.5 mM. PROD and EROD were assayed by measuring the formation of the corresponding hydroxy product in a Perkin–Elmer spectrofluorimeter as described by Krijgsheld and Gram [28]. The concentration of pentoxyresorufin or ethoxyresorufin was 3 μM. ADP, DMNd and HMPAd were estimated by measuring formaldehyde production according to the procedure of Tu and Yang [29] and these demethylase activities were linear at least up to 30 min with 1 mg/mL of microsomal proteins from nasal POOL. Incubations (0.75 mL) were carried out for 30 min at 37° in 100 mM potassium phosphate buffer (pH 7.4) containing 5 mM $MgCl_2$, 0.1 mM EDTA, 0.5 mM $NADP^+$, 10 mM glucose 6-phosphate, 2 units of glucose 6-phosphate dehydrogenase, microsomes (0.4–1.0 mg protein) and substrate (8, 5 and 4 mM for AP, HMPA and DMN, respectively) dissolved in water. Blank tubes containing everything except substrate were run simultaneously and the substrate was added at the end of incubation after protein precipitation by $ZnSO_4$ and $Ba(OH)_2$. Blank values were subtracted from the experimental values. Blanks consisting of substrate and boiled microsomes or immediate protein precipitation by $ZnSO_4$ and $Ba(OH)_2$ (zero time) resulted in lower blank values leading to an erroneous overestimation of these *N*-demethylase activities as previously reported [9].

RESULTS

Preparation of microsomes

The yield of protein and the quality of nasal mucosa homogenate and microsomal fraction, checked by assaying one mitochondrial (cytochrome *c* oxidase) and one endoplasmic reticulum (NADPH–cytochrome *c* reductase) marker enzyme, are reported in Table 2.

The nasal microsomes contained only 4% of the protein of the original homogenate whereas the percentage yield of microsomal protein obtained using human liver is reportedly higher (8–10%) [30]. This probably reflects that only a fraction of hypertrophic nasal samples contained cells with endoplasmic reticulum. It is also possible that nasal cells normally contain less endoplasmic reticulum than liver cells. However, the recovery of nasal mucosa NADPH–cytochrome *c* reductase activity was good (similar to liver) being about 35% of that of the homogenate [30]. In addition, some mitochondrial contamination was detected in nasal microsomes as evident from cytochrome oxidase activity.

Figure 1 shows an electron micrograph of microsomes prepared from the POOL of nasal samples. The microsomal preparation consists primarily of closed double-layer membrane-surrounded microsomal vesicles presumably corresponding to the smooth endoplasmic reticulum.

Nasal mucosa microsomal electron transport enzymes

The microsomal fraction from 10 pooled human nasal mucosa specimens was found to contain cytochrome P450, cytochrome b_5, NADH and NADPH–cytochrome P450 reductase and NADH–cytochrome b_5 reductase (Table 3). The P450 content found was very similar to the value (26 pmol/mg protein) previously reported for two pooled human respiratory tissue samples [9]. In Table 3 the values for the electron-transport enzymes determined in two human livers are also reported for comparison. They are very close to those published earlier [31].

Monooxygenase activities in nasal mucosa and liver microsomes

As shown in Table 4, both the microsomes from nasal POOL and the microsomal fractions of individually prepared nasal samples (numbers 16–25) exhibited a similar content of microsomal protein and similar very weak NADPH–cytochrome *c* reductase and monooxygenase activities towards various xenobiotics such as DMN, 7-ethoxycoumarin and ethoxyresorufin. The AnH, ADP activities and some HMPAd activity were not detected in the individual samples whereas they were measured in the POOL. The pNPH, the DMNd at a DMN concentration of 0.5 mM, and the DENd activities at a DEN concentration of 1 mM were undetectable in the nasal POOL while the DENd, at DEN concentration 50 mM, was found to be 1.0 nmol/mg protein/min, quite close to the values previously reported [10].

The electrophoretic analysis of microsomes from the respiratory samples and the POOL did not show any significant difference (results not shown). The human liver monooxygenase activities which showed values similar to those published earlier [31, 32] were many times higher, except for HMPAd, than those corresponding to nasal tissues.

Phase II enzyme and epoxide hydrolase activities of human nasal mucosa and liver

Several phase II enzymes were investigated both

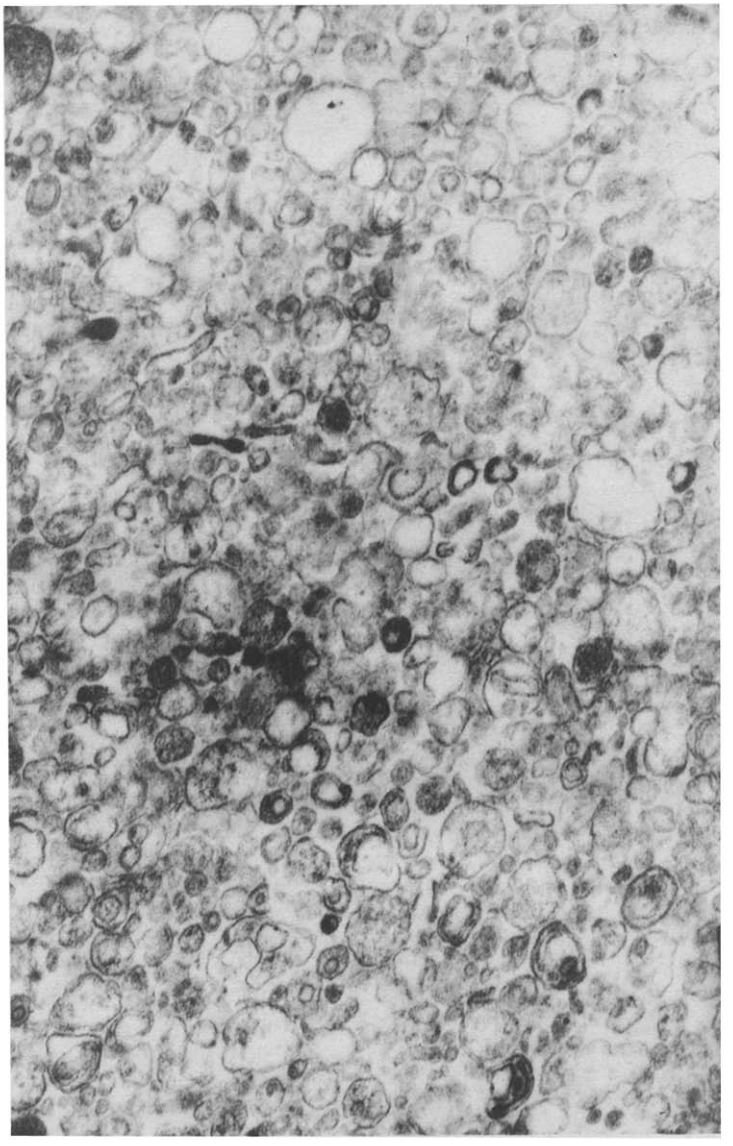

Fig. 1. Electron microscopy of the microsomal fraction obtained from human respiratory nasal mucosa showing smooth vesicles of slightly different sizes. The magnification is 40,000×.

Table 3. Components of the human nasal mucosa P450 monooxygenase system and liver

Parameters	Amount or specific activity	
	Nasal mucosa	Liver
Cytochrome P450 (pmol/mg protein)	24.6	512
Cytochrome b_5 (pmol/mg protein)	5.7	357
NADPH–cytochrome *c* reductase*	23.4	107
NADH–cytochrome *c* reductase*	72	1104
NADH–cytochrome b_5 reductase†	2042	2188

Analysis determined using the microsomal POOL of nasal samples and two human livers. The data reported for the liver are an average of the two samples. The difference between them was 15%.

* Values in nmol cytochrome *c* reduced/min × mg protein.

† Values in nmol of ferricyanide reduced/min × mg protein.

Table 4. Monooxygenase activities in human nasal mucosa and liver microsomes

Patient no.	Protein*	ECOD†	EROD†	AnH‡	NADPH-cytochrome *c* reductase§	HMPAd‖	DMNd‖	APD‖
16	2.43	ND	2.8	<30	3.0	<20	163	<20
17	3.10	<0.2	1.7	<30	2.8	<20	<20	<20
18	3.53	1.9	0.3	<30	3.2	<20	100	<20
19	3.07	1.8	0.2	<30	3.0	93	88	<20
20	3.18	<0.2	0.9	<30	2.8	<20	22	<20
21	2.47	0.5	1.5	ND	8.4	189	113	<20
22	3.01	<0.2	2.2	ND	4.1	<20	<20	<20
23	2.93	1.1	0.5	ND	6.4	74.5	197	<20
24	3.29	1.9	3.1	ND	4.4	<20	95	<20
25	3.14	0.7	2.5	ND	2.5	<20	28	<20
POOL	3.54	2.9	0.5	65	3.4	92	220	65
Liver	11.2	480	102	687	177	240	2045	826

Analysis performed using microsomes from two human livers and ten single and one POOL of nasal mucosa samples. The data reported for liver are an average of the two samples. The difference between them was 22%.

* Expressed as mg/g tissue.

† Expressed as pmol/min/mg protein. Minimal detectable activity was 0.2 pmol/min/mg protein.

‡ Expressed as pmol/min/mg protein. Minimal detectable activity was 30 pmol/min/mg protein.

§ Expressed as nmol/min/mg protein.

‖ Expressed as pmol/min/mg protein. Minimal detectable activity was 20 pmol/min/mg protein.

ND, activity not determined as tissue was not available.

Table 5. Epoxide hydrolase (EH) and phase II enzymatic activities of human nasal mucosa and liver

Patient no.	Cytosolic protein*	EH†	UDP-GT_1‡	BeD†	PrD†	GST†	DT-diaforase†	Carbonyl reductase†
16	10.71	ND	<0.001	12.9	1.15	46.1	2.84	1.64
17	6.28	ND	<0.001	8.9	1.50	127	1.8	0.62
18	6.79	5.6	ND	4.8	0.48	73.5	1.5	0.41
19	8.92	27.2	ND	3.7	1.53	46.9	2.15	0.74
20	8.44	ND	ND	13.6	1.39	119.9	7.7	5.56
21	7.11	33.6	ND	11.7	4.58	85.2	3.92	0.49
22	13.78	13.9	ND	4.4	1.58	42.6	2.75	0.53
23	15.49	26.1	<0.001	6.5	2.1	39.9	7.46	1.9
24	7.28	22.6	<0.001	3.1	1.47	55.8	9.22	2.99
25	11.88	ND	<0.001	4	1.89	31.5	4.11	1.9
POOL	14.5	17.4	<0.001	12.8	2.08	56.6	6.25	1.92
Liver	24.8	35.6	2.4	1.2	3.07	476	0.49	1.85

Analysis performed using 100,000 *g* supernatant or microsomes from two human and ten single and one POOL of nasal mucosa samples. The data reported for liver are an average of the two samples. The difference between them was 17%.

* Expressed as mg/g tissue.

† Expressed as nmol/min/mg protein.

‡ Expressed as nmol/min/mg protein. Minimal detectable activity was 1 pmol/min/mg protein.

ND, activity not determined as tissue was not available.

in microsomal and supernatant fractions from nasal POOL and 10 individual nasal samples (Table 5).

In general the enzymic activities did not vary markedly within the tested samples although fluctuations in some activities could be seen. Whereas the UDP-GT_1 activity was undetectable in all samples investigated, the GST and particularly the microsomal EH activities were relevant and more than 100 times higher than the monooxygenase activities observed for the different substrates of respiratory P450 (see Table 4). The comparison of phase II enzymatic activities of human nasal mucosa and liver, of which UDP-GT_1 and GST rates were comparable to those published [33, 34], showed that the values, except for UDP-GT_1 and GST, were similar in both tissues. However, both DT-diaphorase and BeD activities were somewhat lower in liver than in nasal samples. In addition, whereas in liver DT-diaphorase activity was, as expected [19], markedly lower than carbonyl reductase, in nasal mucosa the reverse held. The presence of both enzymes could protect the respiratory epithelium from the quinones of polycyclic aromatic hydrocarbons which are abundant in all burnt organic material including cigarette smoke and automobile exhausts. The aldehyde dehydrogenases which are known to occur in the nasal respiratory and olfactory regions of rat [35] were found in human respiratory

tissue as well. This enzymic presence could effectively hinder or minimize the noxious effects of reactive aldehydes.

DISCUSSION

The study of drug-metabolizing enzymes in human respiratory nasal mucosa has been hindered by the heterogeneity and the low amount of the specimens and the low levels of monooxygenase activities found in this tissue. The human respiratory epithelium of the nasal cavity contains many distinctive cell types unevenly distributed including ciliated and non-ciliated epithelial cells, globed, cuboidal and basal cells [36]. The samples used in this paper were from hypertrophic tissue of different parts of nose and, of course, from genetically different individuals. However, although the human respiratory samples contained a great deal of connective tissue, the homogenization (using Ultra-Turrax) and the microsomal preparation procedure established for liver could be applied to nasal tissue, as shown by the enzyme markers and electron microscopy analysis, as tools for checking purity and recovery of microsomes.

The data presented herein show that human nasal microsomes contain, although at low levels, all the enzymic components of the monooxygenase system. The human respiratory epithelium contained measurable amounts of P450, at levels similar to those found for the respiratory mucosa of rat and dog but lower than the corresponding value reported for rabbit [2]. The human nasal P450 content was also about double that reported for human lung tissue [30] and about 5% of that of human liver [31, 32]. In spite of such an appreciable P450 concentration, the human nasal tissue showed, compared to human liver, lower monooxygenase activities towards several substrates (except DMN and DEN) even if the activities were expressed per nanomole of P450. This result probably reflects the low amount in the human nasal tissue of certain specific P450 forms along with a low activity of NADPH–cytochrome *c* reductase which was one tenth of that found in whole nasal mucosa of rat [5].

Since the pathology status (hypertrophy) of the nasal samples was the same, as judged by histopathological examination, the observed variations in the enzymic activities were probably due to the heterogeneity of the nasal mucosa specimens as they derived from different parts of the nose. In addition, an inter-individual genetically-based enzymic variation to metabolize xenobiotics among the respiratory mucosa samples might play a role.

In the respiratory nasal mucosa of rat the presence of P450IA, P450IIB1 and P450IIIA have been reported as determined by immuno-histochemical analysis using antibodies raised against hepatic P450s of rat [4]. It is likely that the same or similar P450 isozymes might be present in the respiratory nasal epithelium of humans. However, it should be considered that, apart from P450IIE1, the orthologous P450(s) in human liver appear to be structurally and catalytically very different to those of rat liver [37]. Thus, an identification of nasal P450(s) needs further experiments including the use of antibodies raised against human liver P450(s).

ECOD and EROD activities have been found in the POOL and in nearly all the single human samples tested suggesting that the related P450(s) are widely distributed in the respiratory region. These activities are linked to the P450IA in rats [11, 38] and probably to the same in humans [37]. Other nasal monooxygenase activities such as AnH, HMPAd and APD have been detected only in the nasal POOL and not in the single samples. This finding probably reflects the detection limit of the assay for these activities being two orders of magnitude higher than the assay for the ECOD or EROD (see Table 4). As the P450IIB1 has been detected but not purified from human liver [37] it remains to be established if the nasal HMPAd and APD activities are mainly associated to P450IIB1, as in rat [38, 39], or if other P450 families or subfamilies, as P450IIC and P450IIIA [37], are implied. The P450IIE1 form, although determined in the nasal mucosa of rabbit [40] appears not to be present in the human respiratory epithelium within the detection limits of the assays, since the DMNd, DENd (at low substrate concentrations) and pNPH, all linked to this P450 isozyme [41] have not been detected both in the POOL and any individual human nasal sample. At high substrate concentrations, the DMNd and DENd activities were the highest ones found in the respiratory human tissue and they were 2–5 times higher to those corresponding to human liver when the activities were expressed as the turnover number for total P450.

These results could be due to the occurrence in human respiratory epithelium of unique form(s) of P450(s) not present in liver. In this respect, Ding and Coon [39] have reported a purification of two unique P450 isozymes (P450 NMa and P450 NMb) from nasal microsomes of untreated rabbits both able to catalyse the DEN deethylation [39].

With regard to the non-oxidative and generally detoxicating phase II enzymic activities, they, except UDP-GT_1, were detected in all nasal human samples assayed with few differences among them. This data supports the idea that these enzymes are distributed in all cell types of respiratory human epithelium. A striking finding is the complete lack of UDP-GT_1 activity as determined even in the presence of Triton X-100. As multiple enzyme forms of UDP-GT are known and 1-naphtol is a substrate for UDP-GT_1 (a broadly diffuse UDP-GT form chiefly inducible by 3-methylcholanthrene [42]), it remains possible that other UDP-GT forms might be present in the respiratory tissue of human. High levels of UDP-GT activity, comparable to those of liver, have been reported for nasal mucosa of rat [5, 6] and dog [43] using substrates for UDP-GT_1 such as naphtol or umbelliferon. However, the human lung contains very low activity (10 pmol/mg protein/min) of UDP-GT_1 with 1-naphtol as substrate [33].

Another remarkable result is that EH activity is very high in human nasal tissue and very close to that of human liver in contrast to the nasal mucosa of rat [5], suggesting that metabolically formed or inhaled epoxides could be quickly detoxicated.

Human nasal GST, which was structurally and

immunologically identified as human class π GST is more active in the respiratory tissue than in other human extrahepatic tissues [44]. The measured activity was more than double that of whole rat nasal mucosa [5].

The results taken together show that the activities of the nasal non-oxidative enzymes are similar to those of liver and two or three orders of magnitude greater than the nasal monooxygenase activities. As a consequence, an accumulation of reactive intermediates following bioactivation of inhaled toxicants, involving not in a key role the UDP-GT_1-mediated conjugation route, would not be expected in the nasal respiratory region of humans. However, it should be considered that, generally, phase II enzymes have a relatively low affinity towards xenobiotics. On the contrary, some P450(s) show a very high affinity towards selective substrates. Therefore, metabolic intermediates formed during phase I could not promptly undergo a phase II process. In addition, environmental pollutants present in the air can be concentrated in nasal mucus by factors of 1000–10,000-fold [45] and the role of a single cell type of respiratory nasal mucosa in the balance of activation and detoxication of xenobiotics is not known. Thus, there remains the possibility of bioactivation *in situ* of specific promutagens and procarcinogens.

In conclusion, the results of the present study demonstrate that respiratory sections of human nose contain a wide array of oxidative and non-oxidative enzymes which can be potentially important, particularly if they are inducible, both for overall metabolism of inhaled odorants or volatile xenobiotics and for the susceptibility of different nasal cells to toxicities, although the olfactory region of the nose is expected to exert the major role in both processes.

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