to approximately 50% (*4,5*). *FU* can vary 15-fold among species, with humans in the middle of the range (*12*). This variability is an order of magnitude higher than the reproducibility of the same measurement among laboratories (*4–6,12*). Owing to the lack of human excretion data, fractional excretion values for the rat were used for some congeners. Therefore, our exposure estimates are probably reliable within an order of magnitude.

Exposures for the general population, estimated by the CERHR Phthalates Expert Panel from published data, are in good agreement with our calculated human daily intake estimates based on CDC median values and presented in Table 2. However, the maximal values of excreted monoesters (*1*) indicate that some individual exposures are substantially higher than previously estimated for the general population.

Women of reproductive age appear to be exposed to higher levels of di-*n*-butyl phthalate than are the remainder of the study population. This is particularly evident in the 95th percentile column for *n*-butyl phthalate in Table 2, where the estimated exposure values for women 20–40 years of age are approximately 5 times greater than the corresponding values for the other 192 individuals in the study.

The data reported by Blount et al. (*1*) will certainly lead to further efforts to derive accurate estimates of human exposures based on urinary metabolite levels. In addition, their data lead to several questions that should be addressed in the immediate future; for example:

- What are the sources and circumstances of exposure that result in a higher urinary level of diethyl phthalate metabolites than of the other six phthalates studied?
- What is the evidence for reproductive and developmental toxicity of diethyl phthalate?
- What are the sources and circumstances of exposure that result in some women of reproductive age having higher urinary levels of *n*-butyl phthalate than the remainder of the study population?
- At what levels are humans exposed to other phthalates not included in this study?

It is important that answers to these and related questions be pursued by public health agencies including the NIEHS/NTP.

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Table 3. Comparison of estimated exposures (μg/kg/day) to 97 women aged 20–40 years to the rest of the population (192 individuals)[a] based on extrapolated intake from urinary metabolites (Equation 1) measured by Blount et al. (*1*).

Monoester	Diester	Minimum	Median	95th percentile	Maximum
Ethyl	Diethyl	**0.90**	**13**	**90**	**170**
		< LOD	11	130	320
n-Butyl	Di-*n*-butyl	**0.24**	**1.7**	**32**	**113**
		0.084	1.4	6.5	50
Benzyl	*n*-Butyl benzyl	**0.094**	**1.2**	**4.5**	**7.8**
		0.11	0.78	3.4	29
Cyclohexyl	Dicyclohexyl	< LOD	**0.051**	**0.24**	**0.45**
			0.012	0.25	2.3
2-Ethylhexyl	Di(2-ethylhexyl)	< LOD	**0.71**	**3.8**	**10**
			0.71	3.5	46
n-Octyl	Di-*n*-octyl	< LOD	**< LOD**	**0.65**	**1.5**
			0.015	1.0	13
i-Nonyl	Di-*i*-nonyl	< LOD	< LOD	**3.7**	**7.8**
				1.4	22

[a]Values for women aged 20–40 years in boldface; remaining values are for the rest of population.

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Antitumor Effects of THC

1-Trans-delta^9-tetrahydrocannabinol (THC), the main active component of marijuana, has been shown to exhibit anticancer activity (*1,2*). Galve-Roperh et al. (*1*) reported that intratumoral administration of THC induces apoptosis of transformed neural cells in culture, and also induces a considerable regression of malignant gliomas in Wistar rats and in mice deficient in recombination activating gene 2. These authors suggest that their "results may provide the basis for a new therapeutic approach for the treatment of malignant gliomas." Regarding this interesting finding, we believe it is important to highlight the previous National Toxicology Program's long-term *in vivo* bioassay results that showed definite antitumor activity of THC (*3,4*). This also appears relevant to the current controversy, at least in the United States, regarding the use of marijuana in clinical medicine.

Experimentally, groups of 60–70 male and female rats were administered 0, 12.5, 25, or 50 mg THC/kg body weight (bw), and male and female mice were given 0, 125, 250, or 500 mg THC/kg bw in corn oil by gavage for 104–106 weeks (*3,4*). During this 2-year period, individual animal body weights were reduced compared to controls, although all groups consumed the same amounts of food. More importantly, survival in all THC groups of male and female rats was significantly greater than the controls. For mice, survival was comparable among groups except for the high-dose males. Clinical findings in the THC groups included lethargy followed by hyperactivity, convulsions, and seizures, which occurred typically during and immediately after dosing or handling.

In both rats and mice, no increased incidences of neoplasms were considered related to the administration of THC (*3,4*). In fact, for several organ systems the incidences of background tumors in these strains were actually reduced. The incidences of mammary gland fibroadenomas and uterine stromal

polyps were decreased in THC groups of female rats, as were incidences of pituitary gland adenomas, interstitial cell adenomas of the testis, and pancreatic adenomas in THC-treated male rats. Concerning nonneoplastic lesions in mice, increases of thyroid gland follicular cell hyperplasia occurred in all THC groups, and increases of forestomach hyperplasia and ulcers occurred in THC groups of male mice; yet, no THC-related tumors were observed to progress from these toxic lesions. This common lack of correlation between toxicity and carcinogenicity has long been known (*5–7*). Regarding carcinogenic activity of THC in mice, thyroid gland follicular cell adenomas were somewhat increased only in the lowest THC-dosed group of mice (125 mg/kg); thyroid gland follicular cell adenomas were found in 0/62 control males versus 6/60, 3/61, and 1/57 mice treated with 125, 250, and 500 mg THC/kg bw, respectively, and in 4/60 control females versus 9/60, 3/60, and 1/60 mice); these were considered not significantly related to THC (*3,4*). However, there were significant decreases observed for both benign and malignant liver tumors in male and female mice.

The reduced body weights in these long-term studies may have been contributory to the lowered tumor rates (*8–10*), as most of the reductions in tumor incidences occurred in hormone-controlled organs (*11*). This should not detract from the overall antitumor effects of THC observed in both sexes of these species and strains. Until further studies are accomplished, these reductions in tumor incidences in six organs should be considered caused by or associated with administration of THC.

Our 2-year studies (*3,4*) showed that the observed THC antitumor effects are not confined to the site of injection or administration, and these antitumor effects seem to affect a range of "spontaneous" tumors commonly found in rats and mice. Consequently, the THC-associated antitumor effects are systemically active and are applicable to different tumor types at different organ sites. Again, this lack of specificity might lend credence to the notion that these effects are hormonally mediated and likely related to the observed decreases in body weights. Nonetheless, there were significant reductions in total benign and malignant tumors in all organs combined for both species after THC exposure: in male rats, tumors were found in 98% of controls versus 98, 92, and 90% of groups treated with 12.5, 25, and 50 mg THC/kg bw, respectively; in female rats, tumors were found in 88% of controls versus 82, 86, and 70% of treated groups. Most strikingly, in male mice tumors were found in 73% of controls versus 55, 44, and 30% of male mice treated with 0, 125, 250, and 500 mg THC/kg bw, respectively, and in female mice, tumors were found in 77% of controls versus 52, 43, and 27% of treated groups (*3,4*).

Interestingly, the dose levels used by Galve-Roperh et al. (*1*) were similar to those used in our studies (*3,4*). Their findings also agreed with ours in that THC administration did not affect either food or water intake or hematologic profiles and general clinical chemistry of the animals. Perhaps further animal bioassay studies should be done to learn more about the antitumor effects of THC. For example, animals could be exposed to known carcinogens (e.g., 9,10-dimethylbenz[*a*]anthracene exposure resulting in mammary gland tumors) to determine if THC would block this carcinogenic activity, or transgenic animals (*12,13*) could be used in an attempt to better clarify the mechanism(s) of THC anticarcinogenic activity. More definition of dose-response–antitumor activity relations would be useful, as would studies using paired feeding, to better define the influence of reduced body weight on tumor incidences.

With respect to genetic toxicology (*4*), THC was not mutagenic in *Salmonella typhimurium* strains TA97, TA98, TA100, or TA1535 with or without rat and hamster liver S9 fractions. In cultured Chinese hamster ovary (CHO) cells, THC induced sister chromatid exchanges at the highest dose tested in the presence of S9; at this dose level, cell cycle delay indicative of toxicity was observed. THC did not induce chromosomal aberrations in cultured CHO cells with or without S9 metabolic activation enzymes. *In vivo*, no increase in the frequency of micronucleated erythrocytes was observed in the peripheral blood of male or female mice administered THC by gavage for 13 weeks. Accordingly, THC does not appear to be genotoxic.

Long-term carcinogenesis bioassays have historically and traditionally been used primarily to identify those agents that cause cancer in laboratory animals and hence determine which agents represent a significant cancer risk to humans exposed to these carcinogens (*14–16*). Conversely, these bioassays can and should also be used to identify potential anticarcinogenic agents (*17–19*). Thus, in our studies, rats and mice that received THC for 2 years exhibited body weight reductions, enhanced survival rates, and decreased tumor incidences in several sites, mainly organs under hormonal control. These earlier experimental carcinogenesis results on THC (*3,4*) clearly lend further validity to the notion that cannabinoids may indeed be anticarcinogenic (*1,2*).

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