level in each case. The increase in excretion from these mice when compared with female mice maintained always at +21°C was found to be significant at the 1% level ($P < 0.01$). It was also found that the i.p. injection of 1 ml of distilled water 1 h before dosing with 2-naphthylamine did not result in a significant increase in the excretion of 2-amino-1-naphthol and its conjugates. When the 2-naphthylamine was given by stomach tube the subsequent excretion of 2-amino-1-naphthol and its conjugates was found to be significantly raised ($0.05 > P > 0.02$).

These results are of interest in connection with industrial bladder carcinogenesis. Evidence has been presented that *ortho*-hydroxylated metabolites of aromatic amines (such as 2-amino-1-naphthol) are involved in the induction of bladder tumours by the carcinogenic amines (CLAYSON[3]). The present work suggests that cold and unpleasant working conditions add to the risks of handling the carcinogenic aromatic amines.

2-Naphthylamine appears to be metabolized by two major pathways; N acetylation followed by hydroxylation in the 6 position or hydroxylation in the 1 position without prior acetylation (WILLIAMS[4]). The hydroxylation of 2-naphthylamine to give 2-amino-1-naphthol is carried out by a N.A.D.P.H. dependent enzyme system associated with the microsomal fraction of the liver. The microsomal oxidative enzymes are involved in the metabolism of a wide range of drugs and foreign compounds (BRODIE[5]). The increase in excretion of 2-amino-1-naphthol and its conjugates when 2-naphthylamine is given by stomach tube implies that misleading results could be obtained from studies in drug metabolism in which the drug is administered to experimental animals in this way. The results suggest that the increases in excretion following fright and exposure to cold may be produced by different mechanisms. The increase in excretion during the building work was about 120%, during this period the animals were subjected to both cold and stress whilst the increase in excretion from animals subjected to cold alone or fright (dosing by stomach tube) was of the order of 60%. It may be, however, that the animals were exposed to a greater degree of stress during the building work than during the other experiments.

AXELROD and INSCOE[6] found that the power to hydroxylate acetanilide increased by 70%/g of wet liver and 87%/mg of microsomal protein in rats when they were subjected to cold. They also found that the power to form glucuronides was not affected and N demethylation activity was depressed. There have been few reports of the effects of cold and stress upon drug metabolism by the microsomal oxidative enzymes but the present work appears to be consistent with that of AXELROD and INSCOE.

The mechanism of the process may be associated with a decline in anabolism reducing the rate of oxidation of N.A.D.P.H. (This appears to be an essential co-factor for fat synthesis etc.) There is also evidence that the N.A.D.P.H./N.A.D.P.$^+$ ratio is partly controlled by the rate of glucose 6 phosphate dehydrogenase activity which controls the rate of formation of N.A.D.P.H. by the pentose phosphate pathway. Thus the effects of increased catabolism and decreased anabolism in animals exposed to cold and stress would be to favour N.A.D.P.H. dependent enzyme systems such as the microsomal oxidative enzymes.

Zusammenfassung. An Mäusen wurde festgestellt, dass nach Exponieren in Kälte und Lärm bei dosierter Verabreichung von 2 Naphtylamin mittels Magenschlauch, eine Vermehrung des 2-Amino-1-naphtolderivats in der Exkretion stattfand.

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The Effect of γ-Irradiation on the Amount of 5-Hydroxytryptamine in the Gut and Spleen in the Early Phase after Irradiation

It is known that 5-hydroxytryptamine (5-HT) is a normal constituent of various tissues in the body[1,2], and this substance is also known to be a radioprotective agent[3–6]. It was found previously that the barbiturate sleeping time was prolonged in X-irradiated rats[7] and that this effect of X-irradiation could be partly depressed by methysergide[8], a highly potent and specific 5-HT antagonist. It was therefore decided to study the effect of γ-irradiation and of cysteamine protection on the amount of two typical stores of 5-HT, the gut and the spleen.

Male rats weighing from 190 to 215 g were used in these experiments. γ-Irradiation was administered to rats by a cobalt-60 source. The animals received 900 r with an average of 44 r/min. The animals were sacrificed 24 and 48 h after irradiation. A piece of gut weighing 2 g and the whole spleen were taken out and extracted according to the method of CORREALE[9]. The extracts were assayed biologically on the isolated rat fundus[10] in the presence of atropine (0.1 µg/ml) and antazoline (5 µg/ml). A four point assay was used. In another series of animals, 12 rats in each group, irradiated in the same way and with the same dose, the number of platelets was counted in the peripheral blood in various periods of time after irradiation (24 h, 2, 4, 6, 8 and 14 days). The results are presented in Table I and Table II.

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Table I. The effect of γ-irradiation (900 r) and cysteamine on the amount of 5-hydroxytryptamine in the gut, 24 h and 48 h after irradiation (mean ± standard error in µg/g fresh tissue). The number of experiments is indicated in brackets

1	2	3	4	5	
Controls	24 h after irradiation	48 h after irradiation	24 h after irradiation and cysteamine treatment	48 h after irradiation and cysteamine treatment	P
4.4 ± 0.56 (7)	2.7 ± 0.59 (7)	3.1 ± 0.71 (7)	3.3 ± 0.59 (7)	2.9 ± 0.74 (7)	(1:2) not significant (1:3) not significant (1:4) not significant (1:5) not significant

Table II. The effect of γ-irradiation (900 r) and cysteamine on the amount of 5-hydroxytryptamine in the spleen, 24 h after irradiation

	Controls			γ-Irradiated animals			γ-Irradiated and cysteamine treated animals		
No. of experiment	Weight of the spleen (in mg)	Amount of 5-HT (in µg/whole spleen)	Amount of 5-HT (in µg/g fresh tissue)	Weight of the spleen (in mg)	Amount of 5-HT (in µg/whole spleen)	Amount of 5-HT in µg/g fresh tissue)	Weight of the spleen (in mg	Amount of 5-HT (in µg/whole spleen)	Amount of 5-HT (in µg-g fresh tissue)
1	–	–	4.0	280	4.4	15.6	315	2.1	6.6
2	460	3.8	8.3	260	3.7	14.4	190	1.5	7.9
3	410	2.6	6.8	230	3.5	15.3	180	1.6	8.8
4	480	3.0	6.2	315	4.3	13.8	185	1.7	9.2
5	335	2.3	6.9	225	2.8	12.6	210	1.8	8.6
6	420	2.6	6.3	210	2.9	14.0	270	1.5	5.5
7	425	3.0	7.0	240	2.6	10.9	200	2.9	14.6
8	350	3.2	9.2	275	5.5	19.9	220	3.8	17.3
9	400	1.3	3.2	265	2.5	9.6	335	1.8	5.4
10	630	3.2	5.0	250	3.2	12.7	255	1.6	6.1
Mean ± SE									
434 ± 28		2.7 ± 0.23	6.2 ± 0.57	255 ± 9		3.5 ± 0.30	13.8 ± 0.88	236 ± 17	2.0 ± 0.23	9.0 ± 1.24
(1)		(2)	(3)	(4)		(5)	(6)	(7)	(8)	(9)

P (1:4) < 0.001, P (1:7) < 0.001, P (4:7) not significant, P (3:6) < 0.001, P (3:9) < 0.005, P (2:5) not significant, P (6:9) < 0.001, P (2:8) < 0.05, P (5:8) < 0.005.

The results of the present experiments indicate that γ-irradiation (900 r) did not produce a significant change in the amount of 5-HT in the gut 24 and 48 h after irradiation. This is in contrast with the previous findings of other investigators[11] who have found that 5-HT content in the intestine was decreased 24 h after irradiation. The amount of 5-HT in the spleen 24 h after irradiation, expressed per g of fresh tissue, was significantly increased in γ-irradiated animals and significantly decreased by administration of cysteamine before irradiation. Meanwhile, this increase was not significant if the amount of 5-HT was expressed in µg per whole spleen. This indicates that loss in the weight of the spleen in γ-irradiated rats is not due to deterioration of 5-HT containing elements in the spleen. This dose of γ-irradiation caused a decrease in the number of platelets, 24 h after irradiation, from 767,200 ± 95,000 (standard deviation) in controls to 610,916 ± 86,000 ($P < 0.005$) in irradiated animals and to 622,083 ± 72,000 ($P < 0.005$) in irradiated and cysteamine protected animals. Nevertheless, the observed decrease in platelets was still insufficient to produce a change in the amount of 5-HT in the spleen, as expressed per whole organ. It has been already found that 6 days after irradiation, parallel with a very significant reduction in platelets, the amount of 5-HT in the spleen was significantly reduced[12]. Cysteamine protection was found to decrease significantly the amount of 5-HT in the spleen. The weight loss of the spleen is almost identical in γ-irradiated animals as well as in cysteamine protected animals. Nevertheless, the amount of 5-HT is significantly lower in animals protected by cysteamine than in the irradiated controls.

Résumé. 24 et 48 h après irradiation-γ (900 r) la quantité de 5-hydroxytryptamine dans l'intestin du rat n'a pas changé de manière significative. La quantité de 5-hydroxytryptamine dans la rate du rat, exprimée par g de tissu frais a augmenté. Le prétraitement avec de la cystéamine abaisse nettement la quantité de 5-hydroxytryptamine dans la rate, après l'irradiation.

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