la moyenne des pourcentages maxima de cellules radiolésées est:

dans le rectum: 19,2 ± 1,25% (11 rats) (Tableau III);
dans le vagin: 17,2 ± 1,45% (12 rats) (Tableau IV).

Nous constatons donc une protection à la fois de la muqueuse rectale et de la muqueuse vaginale (radioprotection globale, non spécifique).

C. Après une irradiation intra-rectale de 4 h combinée à une irrigation intra-rectale de cystamine, la moyenne des pourcentages maxima de cellules radio-lésées est:

dans le rectum: 15,8 ± 1,15% (11 rats) (Tableau V);
dans le vagin: 26,7 ± 1,80% (10 rats) (Tableau VI).

Il résulte de ces dernières observations que la muqueuse rectale est électivement protégée par la cystamine administrée localement. En effet, si cette protection était due à une résorption de la cystamine dans le torrent circulatoire, le vagin serait lui aussi protégé comme nous l'avons montré en B; ce n'est pas le cas.

Nous pouvons donc conclure que la cystamine est capable d'exercer un effet protecteur local.

IV. *Conclusions*

Le fait que la cystamine, en application locale, protège le rectum contre une irradiation sans exercer une action d'intensité comparable sur la muqueuse vaginale voisine permet d'envisager pour l'avenir des applications intéressantes, à la radiothérapie des cancers, de certains protecteurs chimiques.

Il est clair que s'il est possible d'accroître la dose-tumeur dans le traitement du cancer du col en évitant, grâce à ces radio-protecteurs introduits sur place, de léser gravement la vessie ou le rectum, un des gros ennuis de la curiethérapie du cancer cervico-utérin sera supprimé ou atténué.

Il ne fait pas de doute pour nous que des techniques de ce genre peuvent être étendues dans d'autres régions du corps, comme le cou, par exemple.

L. Darcis et P. Hotterbeex

Centre anticancéreux près de l'Université de Liège (Belgique), le 7 juin 1957.

Summary

The authors studied the lesions observed in the rectal and vaginal smears of the rat after a local irradiation of the rectum. They observed that an injection of cysteamine before the irradiation strongly diminishes the ratio of damaged rectal and vaginal cells. A local treatment with cysteamine in the rectum before and during the irradiation diminishes only the ratio of damaged rectal cells.

Laboratory Testing of Combined Diphtheria-Tetanus-Pertussis Preparations

Although the efficiency of whooping-cough vaccines has recently been proved in field trials, the problem of combined diphtheria-tetanus-pertussis preparations has been investigated very intensively (Ungar[1], Bousfield and Holt[2], Barr *et al.*[3], Spiller *et al.*[4], Sauer and Tucker[5], Chevé et Zourbas[6]).

Our plan consisted in observing in experimental animals:

(1) the influence of the whooping-cough vaccine on the degree of immunity against diphtheria;
(2) the influence of the whooping-cough vaccine on the degree of immunity against tetanus;
(3) the influence of an addition of diphtheria and tetanus antigens and aluminium phosphate on the degree of immunity against whooping-cough.

Preparations employed and methods of testing.—The following preparations were used:

(1) a combined diphtheria-tetanus purified and adsorbed toxoid (Di-Te);
(2) a combined diphtheria-tetanus purified and adsorbed toxoid with a pertussis vaccine (Di-Te-Per);
(3) a whooping-cough vaccine.

The composition of the various components of these preparations was as follows (per ml): Purified diphtheria toxoid 10 Lf, purified tetanus toxoid 10 Lf, 20,000 millions of pertussis germs, 10 mg of aluminium phosphate. Preservative: merthiolate 1:10,000.

To test the amount of immunity against diphtheria and tetanus, the following methods were used: one stimulans method, two stimulans method, toxic challenge method.

Results obtained.—The influence of pertussis germs added to the combined diphtheria-tetanus preparation on the degree of immunity against diphtheria is presented in Table I.

20×10^7 pertussis germs added to 1 ml of combined diphtheria-tetanus preparation had no influence on the degree of immunity against diphtheria, according to the one stimulans method, since there was no significant difference in the average amount of A.U. in the serum of the immunized guinea-pigs. The two stimulans method, however, showed a significant difference in favour of the combined diphtheria-tetanus-pertussis preparation. The results of the toxic challenge method also appeared to be in favour of the combined diphtheria-tetanus-pertussis preparation. According to this method, it appears that an addition of pertussis germs enhances the immunity against diphtheria.

The influence of pertussis germs added to the combined diphtheria-tetanus preparation on the degree of immunity against tetanus is presented in Table II.

20,000 million pertussis germs added to 1 ml of combined diphtheria-tetanus preparation did not influence the degree of immunity against tetanus. A comparison of the degree of immunity against tetanus between the guinea-pigs vaccinated with the combined diphtheria-tetanus preparation, and others vaccinated with the combined diphtheria-tetanus-pertussis preparation, showed no significant difference in the degree of immunity either by the one stimulans method, the two stimulans method or the toxic challenge method.

[1] J. Ungar, Proc. Roy. Soc. Med. *47*, 355 (1954).

[2] G. Bousfield and L. B. Holt, Med. Officer *92*, 289 (1954).

[3] M. Barr, A. T. Glenny, and N. R. Butler, Brit. med. J. *II*, 635 (1955).

[4] V. Spiller, J. M. Barnes, L. B. Holt, and D. U. Cullington Brit. med. J. *II*, 639, 663 (1955).

[5] L. W. Sauer and W. H. Tucker, Amer. J. publ. Health *44*, 784 (1954).

[6] J. Chevé and J. Zourbas, *Atti del II. Congresso Internazionale di Standardizzazione Immunobiologica*, p. 121 (1956).

Table I: Degree of immunity against diphtheria

	One stimulans method		Two stimulans method		Toxic challenge method		
	Di-Te	Di-Te-Per	Di-Te	Di-Te-Per	Dilution	Di-Te Total death	Di-Te-Per Total death
Arithmetic mean of A.U.	3·44	3·064	4·47	6·38	1/100	1/20	1/20
Mean of log. A.U.	0·4856 ± 0·242	0·3714 ± 0·372	0·620 ± 0·101	0·8086 ± 0·104	1/200	19/20	19/20
Geometric mean of A.U.	3·05	2·35	4·17	6·41			
Difference between samples	$0{\cdot}5 > P > 0{\cdot}4$		$P < 0{\cdot}001$				

Table II: Degree of immunity against tetanus

	One stimulans method		Two stimulans method		Toxic challenge method		
	Di-Te	Di-Te-Per	Di-Te	Di-Te-Per	Dilution	Total death	Total death
Arithmetic mean of A.U.	2·57	2·66	18·82	17·34	1/50	0/20	0/20
Mean of log. A.U.	0·10 ± 0·661	0·1616 ± 0·869	1·2066 ± 0·169	1·157 ± 0·199	1/100	8/20	8/20
Geometric mean of A.U.	1·251	0·689	16·10	14·30			
Difference between samples	$0{\cdot}5 > P > 0{\cdot}4$		$0{\cdot}5 < P > 0{\cdot}4$				

The pertussis vaccine with addition of aluminium phosphate and diphtheria and tetanus toxoids proved significantly better than the pertussis fluid vaccine ($0{\cdot}01 > P > 0{\cdot}001$). In additional experiments, the same vaccine batch to which only aluminium phosphate was added proved also better than the fluid vaccine.

Conclusion.—Laboratory tests of a combined diphtheria-tetanus-pertussis preparation consisting of 30 Lf of purified diphtheria toxoid mixed with 10 Lf of purified tetanus toxoid and 20,000 millions of *H. pertussis* germs, adsorbed on 10 mg of aluminium phosphate, showed that the composition of the preparation was well balanced and that efficient immunity was conferred at the same time against diphtheria, tetanus and whooping-cough.

Acknowledgement.—The author is indebted to J. BENKOVIĆ, LJ. MANDIĆ, N. KÖHLER, and N. ŠKARICA for their help in this work.

D. IKIĆ

Institute for the Control and Research of Immunobiological Substances, Zagreb (Yugoslavia), July 9, 1957.

Zusammenfassung

In Laboratoriumsuntersuchungen wurde der gegenseitige Einfluss der einzelnen Komponenten des auf Grund purifizierter Anatoxine zubereiteten und auf Aluminiumphosphat adsorbierten Mischimpfstoffes gegen Diphtherie, Tetanus und Pertussis festgestellt.

Pyridoxin and the Acute Toxicity of Isoniazid and Other Acid Hydrazides in Guinea Pigs[1]

In an earlier communication from this laboratory[2], it was demonstrated that the acute toxicity produced in guinea pigs with acid hydrazides is different from that of free hydrazine compounds. It was further shown that the toxicity of the acid hydrazides alone responded to the administration of pyridoxin. Isoniazid and cyanacetic acid hydrazide were the two substances investigated in that study, and this article represents the results of further experimentation with the hydrazides of pyridine carboxylic, benzoic and acetic acids.

The methods used have been described earlier[2]. A total of 273 guinea pigs were used in this experiment. All compounds, dissolved in water, were administered by intraperitoneal injection. Pyridoxin, when given, was injected immediately before by the same route. The animals were then observed for a period of 20 h for signs of toxicity and the survival time noted.

The results of these experiments show that the central stimulatory action of isoniazid could also be produced by other acid hydrazides whether they are derived from a pyridine carboxylic acid, benzoic acid, or an aliphatic carboxylic acid, such as acetic acid, although the degree of toxicity of these compounds was variable.

From the results presented in Table I, it is seen that pyridoxin exerted a protective effect against the toxicity of all the hydrazides studied. It seems, therefore, that there exists no specific metabolite-antimetabolite relationship between pyridoxin and isoniazid alone. The possibility was considered that pyridoxin simply exerts its effect by forming a hydrazone which may be excreted, as postulated by BIEHL *et al.*[3]. In order to investigate this problem, ethylidene-isoniazid representing the hydrazone of isoniazid and acetaldehyde, was studied for its toxicity. This substance, it was presumed, cannot chemically interact with pyridoxal as the free end of the hydrazine group is bound. Results indicate that ethylidene-isoniazid was less toxic than isoniazid, and only at higher doses produced symptoms of toxicity, which were modified by pyridoxin. It is possible that ethylidene-isoniazid was hydrolysed within the body to yield free isoniazid, which then produced the toxic effect.

[1] The authors are indebted to J. R. Geigy S.A., Basle, for the preparation of the acid hydrazides studied.

[2] M. O. TIRUNARAYANAN and W. A. VISCHER, Exper. *12*, 291 (1956).

[3] J. P. BIEHL and R. W. VILTER, Proc. Soc. exp. Biol. Med. *85*, 389 (1954).