yellow N-*o*-nitrophenysulfenyl-L-alanine methyl ester (III); m.p. 75.5–76.5°; $[\alpha]_D^{27.2}$ — 72.0° (c 1, dimethylformamide). The addition of hydrazine hydrate to a methanolic solution of the Nps-ester III[7] formed within minutes crystalline N-*o*-nitrophenylsulfenyl-L-alanine hydrazide (IV); m.p. 183–185° (with effervescence); $[\alpha]_D^{27.1}$ — 51.0° (c 1, dimethylformamide). Solid *p*-nitrophenyl trifluoroacetate[8,9] was mixed with a solution of the Nps-hydrazide IV and triethylamine in dimethylformamide to afford N^1-*o*-nitrophenylsulfenyl-N^2-trifluoroacetyl-L-alanine hydrazide (V); m.p. 203–205°; $[\alpha]_D^{26.7}$ — 43.0° (c 1, dimethylformamide). A solution of the Nps-Tfa hydrazide V[7] and standardized hydrogen chloride in ethyl acetate[10] gave N^2-trifluoroacetyl-L-alanine hydrazide hydrochloride (VI) as a hydroscopic, colorless gel. The Tfa-hydrazide salt VI was coupled to N-benzyloxycarbonyl-L-alanine (VII)[11] in the presence of triethylamine with the aid of either N,N'-dicyclohexylcarbodiimide[12] or 1-ethyl-3-(3-dimethylamino)propyl carbodiimide hydrochloride[13] in methylene chloride or chloroform solutions to yield oily N-benzyloxycarbonyl-N^2-trifluoroacetyl-L-alanyl-L-alanine hydrazide (VIII). The Z-Tfa dipeptide hydrazide VIII[7] in methanol was refluxed with dilute, aqueous sodium hydroxide so as to cleave the Tfa group and to form N-benzyloxycarbonyl-L-alanyl-L-alanine hydrazide (IX), identical with an authentic sample[14–16]. Most importantly, this overall sequence demonstrates that the N^2-Tfa moiety can be carried through a series of typical peptide reaction steps, including introduction in a neutral solvent, hydrolysis of a second and different acid labile primary amino protecting group, and then removal under basic conditions.

Alternatively, a solution of N-benzyloxycarbonyl-L-alanine hydrazide (X)[14] and trifluoroacetic acid in tetrahydrofuran was treated with N,N'-dicyclohexylcarbodiimide to yield quantitatively N^1-benzyloxycarbonyl-N^2-trifluoroacetyl-L-alanine hydrazide (XI); sublimable at 165°/0.3 mm.; m.p. 182–185°; $[\alpha]_D^{26.7}$ — 13.0° (c 1, dimethylformamide). Saponification of the Z-Tfa hydrazide XI with dilute, alcoholic sodium hydroxide at room temperature returned the starting Z-hydrazide X. Hydrogenation of compound XI in methanol with excess acetic acid in the presence of 10% palladium on carbon catalyst furnished oily N^2-trifluoroacetyl-L-alanine (XII). If the acetic acid was omitted, the product was a mixture of N^2-Tfa alanine XII and a second material, possibly N^1-trifluoroacetyl-L-alanine hydrazide (XIII). The isomeric Tfa-hydrazide XIII could have been derived from a N^2-Tfa → N^1-Tfa transfer.

The preparation of N^1-*t*-butyloxycarbonyl and related N^1-Schiff base amino acid hydrazides[17], followed by conversion to the corresponding N^2-Tfa derivatives, would be a logical extension of the work reported here. Cleavage of the N^1-group under mild acidic conditions will produce N^2-Tfa acyl hydrazides that can be routinely coupled to various N^1-acyl amino acids. Attention is called to the point that the hitherto elusive ω-nitroarginyl peptide hydrazides may be synthesized in this fashion, as direct treatment of the nitroguanido group with hydrazine leads to undesirable results[18–21]. These various suggestions are under active investigation[22].

Zusammenfassung. Die Bildung von Z-N^2-Trifluoroacetyl-Aminosäure-Hydraziden wird beschrieben. Die Anwendungsmöglichkeiten dieser Verbindungen in der Peptidsynthese werden durch mehrere Beispiele demonstriert.

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[22] The authors are indebted to the National Science Foundation for Grants No. GB-587 and GP-3888, which supported this study.

Chromosomal Rearrangements Induced in *Drosophila* Salivary Gland Chromosomes by an Acridine Half Mustard

The monofunctional nitrogen mustard derivative of acridine, 2-methoxy-6-chloro-9-[3-(ethyl-2-chloroethyl) aminopropylamino] acridine dihydrochloride (ICR-170), is an efficient mutagen in *Drosophila*[1–3], *Neurospora*[4] and *Salmonella*[5], in addition to its possessing nucleotoxic activity comparable to bifunctional alkylating agents[6]. Mutations induced by ICR-170 at the dumpy locus in *Drosophila*[1] and *ad-3* region in *Neurospora*[4] have been found not to involve chromosomal deletions. Evidence of ICR-170-induced (a) nonsense, non-complementing mutations in *Neurospora*[4] and (b) reversions of frameshift mutations of *C* gene of the histidine operon in *Salmonella*[5] suggests this chemical may be causing mutations of frameshift type in both *Neurospora* and *Salmonella* and possibly

also base pair substitutions in *Neurospora*. In tests on *Vicia faba* chromosomes, ICR-170 has been shown to be a potent chromosome-breaking agent inducing chromatid and isochromatid deletions, and subchromatid, chromatid and chromosome exchanges[7]. The experiments reported here show that inversions result in the salivary gland chromosomes of *Drosophila* after treatment with ICR-170.

Freshly laid eggs of Oregon-K strain were plated on the ICR-170 medium. This medium was prepared by mixing equal volumes of 200 µg of ICR-170 per ml of distilled water and basic medium. Basic medium was composed of agar (3%), yeast (10%), glucose (10%), propionic acid (0.4%) and water (100 ml). For the control, eggs were plated on the basic medium diluted with distilled water (1:1). Third instar larvae from control and experimental media were harvested for studying salivary gland chromosomes. The treatments were conducted in darkness to eliminate the possibility of the photodynamic action of the acridine nucleus. Salivary glands were excised in saline water and squashed immediately in aceto orcein. (ICR-170 was the kind gift from Dr. H. J. CREECH, of the Institute for Cancer Research, Philadelphia, USA.)

4 spontaneous and 52 ICR-170-induced inversions were recovered in 500 cells scored for control and treated larvae. The inversion frequency per cell works out at 0.008 and 0.104 for control and treated larvae respectively. Most of the ICR-170-induced inversions were complex. The concentration of ICR-170 used in these experiments yielded 5.8% sex linked lethals.

ICR-170 has 2 components: an acridine nucleus and a monofunctional nitrogen mustard side chain. Mutagenicity of acridines in bacteriophages is related to acridine intercalation[8] and that of alkylating agents to reaction of agents with nitrogenous bases of DNA, primarily by alkylation of guanine at the N7 position[9]. Whereas the alkylating agents are potent chromosome-breaking agents[7,10–12], the acridines have a milder action and that only under photodynamic conditions[13]. From the specificity of reversions of frameshift, amber, ochre and missense types of mutations in the *his C* gene of *Salmonella*, it is suspected that ICR-170, at low concentrations, may be acting only by the acridine nucleus and not by the alkylating side chain[6]. Chromosome aberration induction studies have led us to suspect that alkylating chain is necessary for its induction of lesions in chromosomes[7].

Zusammenfassung. Bei Kontrolle der Speicheldrüsen ergibt sich, dass die Acridinbehandlung bei *Drosophila* Komplexinversionen erzeugt.

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Elective Localization of ^{3}H-Corticosterone in Mast Cells

Following glucocorticoid treatment, the number of mast cells increases[1–3]; this result, however, is not merely quantitative but also qualitative, leading to the accumulation of the granules of the mast cells, which finally disintegrate[4]. At this phase the observable number of mast cells recedes. This physiological control of the formation and disintegration of mast cells effected by glucocorticoids seems fairly obvious; nothing is known, however, of the cellular mechanism of this control.

Mast cell transformation can be followed easily in subcutaneous connective tissue[4] but still better in the lymphatic organs[1,2,5–7]. Here some cells – reticular cells and lymphoblasts – transform into mast cells. It is not known whether this is in any way connected with the cortisone-cortisol transformation observable in the lymphatic organs. As is known from the experiments of DOUGHERTY et al.[8], the system of 11β-hydroxydehydrogenase is present in the lymphatic organs and, during a prolonged cortisone treatment, activity of the enzyme significantly increases in the thymus. This can be hypothetically connected with the simultaneous appearance of this massive mast cell formation.

Since the adrenal cortex of rats and mice produces first and foremost corticosterone[8], the adult male BALB/C mice used in our experiments were given corticosterone-1,2-^{3}H. The specific activity of corticosterone was 945.9 mCi/m*M*. Labelled corticosterone was diluted with a non-labelled one, and after its suspension with Tween 80 was injected i.p. Thus 1 mg/30 g of corticosterone was given, one tenth of which was active. The activity given was 5 µCi/g body weight. 6 animals were treated in this manner. Material was taken from 2–2 animals 1, 3 and 6 h following treatment. The apex of

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