terminal ileum[7,8] and œsophagus[9] of the guinea-pig. Recently, CHRISTENSEN and DANIEL[10] observed that noradrenaline produced contractions of the strips from the lowest centimeter of the cat oesophagus. This contractile response was opposed by tolazoline and atropine but not by propranolol. These authors were of opinion that adrenergic α-receptors in the cat oesophagus are excitatory. Since according to LANDS et al.[11] both types of catecholamine receptor (that is α- and β-receptors) are present in intestine, low doses of A stimulate either β-receptors only or both α- and β-receptors. In the latter case it is possible that the ratio of α- and β-receptors distribution in various segments of intestine is responsible for the different degrees of relaxation of smooth muscles due to general prevailing amount of β-receptors against α-receptors in intestine and their varying distribution. Further experiments to elucidate the phenomenon observed are in progress.

Zusammenfassung. Adrenalin bewirkt eine triphasische Reaktion am isolierten Rattenileum. Die Kontraktionsphase beruht wahrscheinlich auf Stimulierung der α-Rezeptoren.

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Immunopharmacologic Activity of 1-Aminocyclopentane-1-Carboxylic Acid

1-Aminocyclopentane-1-carboxylic acid (ACPC) is a non-metabolizable, unnatural amino acid lacking an α-hydrogen atom. First synthesized in 1911[1], it was not tested for biological activity until the 1950's, when it was reported to inhibit growth of various experimental tumors and be actively concentrated in malignant cells[2–4]. With the exception of some activity in multiple myeloma, ACPC has been essentially ineffective as an antitumor agent in humans[5]. Its mechanism of action is unknown despite intensive investigation. Aminoaciduria results after oral administration in man, but loss of amino acids is probably not responsible for antitumor activity[6]. ACPC is inactive as an amino acid antagonist in bacterial systems; however, it antagonizes incorporation of valine into proteins in the rat, possibly by preventing attachment of valine to S-RNA[7]. The toxic effects of ACPC can be prevented in chickens by valine[8] and in *Escherichia coli* by methionine and valine[9]. One of the initial signs of toxicity is body weight loss, which can be prevented by force-feeding[10]. Other studies show that ACPC does not inhibit enzymatic oxidation or transamination of amino acids[11] and is not incorporated into protein[12]. The long half-life of ACPC, 20–30 days in rodents and 2–3 days in monkeys and humans[13] is probably due to extensive renal reabsorption and lack of metabolism. These biological characteristics have led to its use as a model for the study of amino acid transport[14].

The immunosuppressive properties of ACPC became evident through our observation that it could suppress experimental allergic encephalomyelitis (EAE) in rats in a dose-related manner[15] (Figure 1). This laboratory-induced immunopathy, a prototype syndrome of cellular hypersensitivity reactions, is useful for detecting and evaluating immunosuppressive agents[16,17]. In this procedure, animals are evaluated for gross hind limb paralysis 14 days after administration of an encephalitogenic emulsion in the hind foot-pad[16]. Study of a series of ACPC analogs indicated that the 5-membered ring is essential for immunosuppressive activity in EAE. Alkylamino substitutions greater than methyl decrease activity. In addition to preventing paralysis, ACPC prevents or suppresses other sequela of EAE, such as reduction of body and stress organ weights, hematologic changes, paw swelling and histopathologic lesions of the brain and spinal cord. Methionine at 5 times and valine and leucine each at 28 times the dose of ACPC failed to prevent its immunosuppressive action in EAE. ACPC likewise suppressed EAE in rabbits and guinea-pigs. At similar doses it is effective in other models of cellular hypersensitivity; for example, it reduces a dinitrochlorobenzene-induced contact dermatitis in guinea-pigs and significantly increases the rejection time of skin grafts in mice (BALB/C to CBA/2).

ACPC is also effective in decreasing circulating antibody responses, including the primary type obtained in the mouse by injecting sheep red blood cells or *Salmonella*

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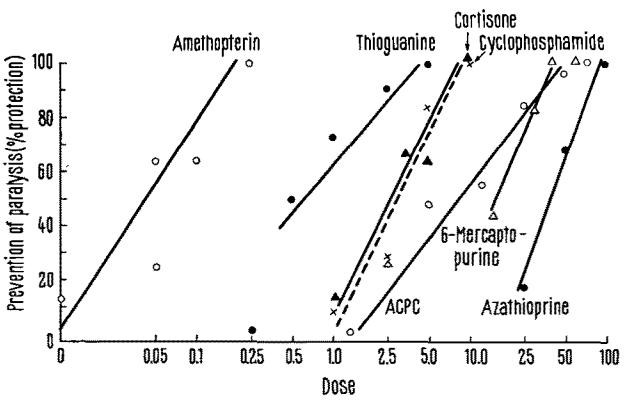

Fig. 1. Effect of immunosuppressive agents on EAE in Lewis strain rats. Animals were evaluated for positive signs of EAE 14 days after administration of an encephalitogenic emulsion of isologous spinal cord in complete Freund's adjuvant. Animals were scored as positive if they exhibited any or all of the signs of EAE such as, hind limb muscular weakness, ataxia, irregular gait, complete paralysis including fecal impaction, urinary incontinence and abdominal wall flaccidity. Compounds were begun on day of antigen administration and given orally as mg/kg on alternate week days for a total of 6 doses, except cortisone acetate, which was administered daily s.c. 5 days/week for a total of 10 doses. Toxicity was not encountered at the doses used. Per cent protection was calculated by comparison with control animals done simultaneously with each drug group. Controls averaged 90–100% paralysis by day 14. Each point represents data on at least 12 rats.

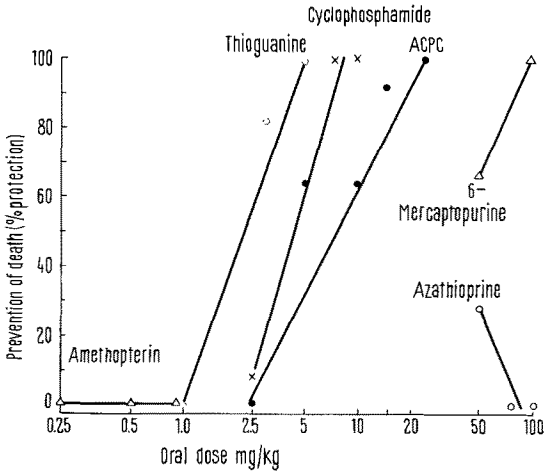

Fig. 2. Effect of immunosuppressive agents on anaphylactic shock in Sprague-Dawley rats. Rats were sensitized with 25 mg ovalbumin and 0.5 ml *H. pertussis* vaccine given i.p., and challenged i.v. 13 days later with 1 mg ovalbumin. Prevention of death was calculated 24 h after antigen challenge by comparison with controls done simultaneously. Anaphylactic deaths in controls averaged 50–60%. Drugs were begun on the day of antigen administration and then given on alternate days for a total of 7 doses. Azathioprine caused deaths in 5 out of 10 animals at 75 mg/kg and 3 out of 10 at 100 mg/kg, while 6-mercaptopurine at 100 mg/kg killed 5 out of 10 rats. These deaths occurred prior to antigen challenge on day 13. Each point represents data on 10 rats except those in which toxic deaths occurred.

typhosa, the same type obtained in the rabbit with bovine serum albumin or globulin, and the secondary or tertiary type obtained in monkeys by challenge with influenza vaccine. Anaphylactic shock is prevented in the rat but not the guinea-pig, with doses equivalent to those used in EAE (Figure 2).

In contrast to the steroidal and non-steroidal anti-inflammatory drugs, ACPC is ineffective against the edematous or early permeability as well as the humoral, neural and biochemical phases of the inflammatory response. Perhaps through their effects on the rapidly proliferating cells responsible for the more chronic stages of inflammation, immunosuppressive agents can exert an anti-inflammatory effect in adrenalectomized animals or in humans without inhibiting the immune response, lowering the leucocyte count or depressing the bone marrow[18]. ACPC generally resembles other immunosuppressive agents in this respect and is active versus the more chronic cellular types of inflammation in rats at doses having little hematologic effects. The models tested include various forms of granuloma (induced by croton oil and cotton) and adjuvant arthritis. ACPC is effective in decreasing primary and secondary lesions in rats with previously established adjuvant arthritis. In contrast to other immunosuppressive agents[19], ACPC is effective in preventing sulfanilamido-indazole-induced periarthritis in rats.

ACPC is inactive in the nephrosis induced by anti-kidney antiserum and in the dextran anaphylactoid reaction in rats and passive cutaneous anaphylaxis in guinea-pigs. The compound has no acute effects on the cardiovascular, renal or central nervous systems of various species at therapeutic doses. Unlike purine, or folic acid antagonists or steroids, ACPC is non-toxic to human embryonic fibroblasts in tissue culture. In rodents, sustained therapeutic effects are obtained after just single doses of drug, undoubtedly due to the long half-life in this species. Depression of the leucocyte count is not seen in any studies using therapeutic doses of ACPC.

The data so far acquired appear to dispute any peripheral mechanism and to indicate that ACPC acts by inhibiting the proliferation of immunologically active cellular lymphoid elements. Its indicated spectrum of immunopharmacologic activity suggests that it may be useful in homotransplantation and various immunological diseases[20].

Zusammenfassung. Es wird gefunden, dass 1-Aminocyclopentan-1-Carboxylsäure (ACPC) die Bildung humoraler und zellgebundener Antikörper in verschiedenen Tierarten hemmt. ACPC ist überdies fähig, die chronische zelluläre Phase des Entzündungsprozesses zu unterdrükken. ACPC hat keine peripher pharmakologischen Angriffspunkte und wirkt offenbar nur auf zelluläre, lymphoide Elemente, welche für Entzündung und Allergie verantwortlich sind. Aufgrund dieser Eigenschaften dürfte ACPC bei Homotransplantationen und immunologischen Krankheiten Verwendung finden.

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