cator paper) and buffered at *p*H 7·8 with 0·5 ml. of 0·2 *M tris* buffer. The suspension formed is incubated with 0·2 mgm. of crystalline trypsin (Worthing Labs.), for 30 min. At the end of this period the enzyme in the clear digest is inactivated by adding 5 ml. of boiled alcohol. After immersion of the tubes for 5 min. into a water-bath adjusted at 70° C. the liquid is quantitatively transferred to a 125-ml. round-bottom flask and evaporated with reduced pressure to dryness. The dry material is suspended in 4 ml. of saline and assayed for bradykinin in a guinea pig ileum preparation using the 2 × 2 bioassay described by Rocha e Silva[4].

The bradykinin content does not change significantly if the blood is treated 2–4 hr. after collecting or if the dry digest is assayed after keeping it in an evacuated desiccator 12–24 hr.

The standard used for comparison was a preparation containing 3 units of bradykinin/mgm.; one unit of this preparation was equivalent to 0·5 μgm. of synthetic bradykinin (prepared by the Sandoz Labs.) according to the procedure of Boissonnas *et al.*[5]. One unit of bradykininogen is defined as the amount of the precursor able to liberate one unit of standard bradykinin or 0·5 μgm. of synthetic bradykinin.

Table 1 shows the concentration of bradykininogen found per ml. of blood plasma of man and other mammals measured according to the method described above.

Table 1. BRADYKININOGEN (UNITS/ML. PLASMA)

	Average	Range
Man	21·2	14·8–28·5
Ox	28·9	23·7–33·9
Horse	18·9	17·6–19·7
Sheep	12·7	12·0–13·8
Pig	8·1	5·5–11·6
Dog	12·3	12·0–13·0
Rabbit	20·1	17·5–22·7
Guinea pig	17·7	15·8–20·0
Rat	4·0	2·5– 5·0

Average of determinations made on the plasma of 10 men and 5 women (normals); other species, average of 3 animals.

The method described is being used in this laboratory for the examination of conditions expected to alter the bradykininogen content of plasma in several types of experimental shock, surgical conditions and pregnancy.

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Biological Spectrum of Some New Tuberculostatic 1,3,4-Oxadiazolones with Special Reference to Cross-Resistance and Rates of Emergence of Resistance

THE tuberculostatic activity of 5-(4-pyridyl)-1,3,4-oxadiazolone (*S* 57) has been repeatedly confirmed[1–3], and complete cross-resistance with isonicotinic acid hydrazid (isoniazid) in laboratory tests using *Mycobacterium tuberculosis* is reported by all workers in this field. It was, therefore, a little surprising to find that, whereas isoniazid is practically inactive against human leprosy, but active against rat murine leprosy, *S* 57 is highly active against both animal and human leprosy[4–6].

Although isoniazid and *S* 57 show complete cross-resistance in laboratory tests, yet if *H* 37 *Rv* is cultured in the presence of a concentration of isoniazid just sufficient to reduce, but not completely inhibit, growth, and a concentration of *S* 57 one-tenth of that required for growth inhibition, the rate of emergence of resistance to isoniazid is substantially reduced, showing that some difference in the biological spectra of isoniazid and *S* 57 is probably present[7,8]. This observation might have some bearing on the unexpected activity of *S* 57 in human leprosy, the more unexpected since it contains, in contrast to most leprostatics, no sulphur in the molecule.

The low toxicity and low local irritant properties of *S* 57 led to the extension of the above observations to the salicylate and *p*-amino-salicylate class of substance which shows reduced therapeutic usefulness owing to local irritant properties[9]. Conversion of the free carboxy group to the corresponding 1,3,4-oxadiazolones yields compounds possessing similar pharmacological spectra to the original salicylate and *p*-amino-salicylate, but showing, in fact as predicted, reduced toxicity and local irritant action. Thus 5-(2′-hydroxyphenyl)-1,3,4-oxadiazolone is an active anti-inflammatory, anti-pyretic and analgesic agent of low irritant properties and toxicity. 5-(2′-hydroxy-4′-aminophenyl)-1,3,4-oxadiazolone (*WS* 127) shows a similar tuberculostatic spectrum and order of activity as *p*-amino-salicylic acid, but is considerably more stable chemically. Here again complete cross-resistance to various strains of *Mycobacterium tuberculosis* has been observed in laboratory tests using strains resistant to *p*-amino-salicylic acid. Yet, on culturing *H* 37 *Rv* in the presence of both *p*-amino-salicylic acid and *WS* 127 at slightly subliminal concentrations the rate of emergence of resistance to both substances is substantially reduced compared with findings obtained using *p*-amino-salicylic acid or *WS* 127 individually on the same strain. The same phenomenon has been noted using *p*-amino-salicylic acid and 5-(2′-hydroxy-4′-aminophenyl)-1,3,4-oxadiazol-2-thione.

Thus, the laboratory cross-resistance tests commonly used apparently miss some of the finer details of activity, at least in this class of substance, which may be necessary to predict therapeutic effectiveness and rates of emergence of resistance both in animals and man.

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