REVIEW ARTICLE

THE RAUWOLFIA ALKALOIDS

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OVER the past two or three decades much attention has been paid by clinicians to the treatment of hypertensive disease. This has resulted in the introduction and trial of a number of drugs, including, amongst others, thiocyanates, nitroprussides, azides, hydrazinophthalazine (hydrallazine), veratrum, the ganglion blocking agents, ion exchange resins, barbiturates and the various extracts and alkaloids of *Rauwolfia serpentina* Benth.

The introduction and use of *Rauwolfia serpentina* is of particular interest; not only has it intriguing pharmacological properties, but it represents something of a return to the use of vegetable drugs, in an age which is becoming increasingly devoted to the use of synthetic chemicals in medicine.

Although rauwolfia may ultimately not survive to take its place alongside the solanaceous drugs, the cardiac glycosides and opium, a study of its properties seems likely to lead to a clearer understanding of the complex mechanism underlying hypertensive disease in man. In this way it may establish itself firmly in the history of medicine. This may seem to be a jaundiced view to take of a new drug which is being widely used and widely studied. Many natural products have proved to be the starting points for further investigation and have yielded place to newer drugs with structures based upon the originals. A study of the literature indicates that this may be the fate of rauwolfia.

HISTORICAL

Rauwolfia serpentina under many names has been used for centuries in folk medicine in India. There are also reports of its use in Burma, Malaya and Java. It is variously called *sarpagandha* (Sanskrit)[1], *Chota-chand* (Hindi)[1,3,4], *chandra* or *chota-chand* (Bengali)[1], *dhan barua* or *dhan marua*, *pagla-ka-dawa* (Bihari)[1,2], *chota-chand*, *chandra*, *karavi*, *harkai* (Bombay)[1,2,4], *harkaya* (Marhatti)[1,2], *atalagandhi* or *patala garuda* (Telugu)[1,2], *Chuvana-avilpori* (Malay)[1,2], *dhannerna ordhan-barua* (Oriya)[2], *covannamiloori* (Tamil)[2], *chandrika* (Sanskrit)[2,3], *Tsjovanna Amelpodi* (Malealie, Malabar Coast), *chivan amelpodi* (Tamil), *Ratu-eka-weriya* (Cyng.), "*Ophioxylon of Serpents*", and *Ophioxylon serpentinum* (Lin.)[3], Trease and Evans[5] point out that the plant is mentioned in an ancient Hindu manuscript of 1000 B.C. In the second century A.D. it is to be found mentioned in Charaka's works[2] as *Sarpagandha*. Ainslie[3] describes the use of the root either as a powder or in the form of a decoction in snake bites and scorpion stings. He mentions also its use as a febrifuge, an anthelmintic, a stomachic and in obstetrics "to promote delivery in tedious cases". He seems to have been in some doubt about its utility

in snake bites, having "invariably trusted to the prompt use of Madeira wine and generally with success". Ainslie[3] gives references to earlier works. Dymock[4] indicates that the root was also used in dysentery: "In Bombay most of the labourers who come from the southern Concan keep a small supply of the root which they value as a remedy in dysentery and other painful affections of the intestines". The Pharmacopœia of India (1868)[6], and other publications[7–15], describe the use of the plant in folk medicine.

Distribution

Rauwolfia serpentina is found in India, Ceylon, Burma, Siam, Malaya and Java. Vakil[2] mentions its occurrence in the Himalayas, Assam, Pegu, Tennasserim, Bihar and the Deccan peninsula.

Botany and Pharmacognosy

The genus *Rauwolfia* belongs to the Apocynaceæ. The genus is named after Leonhard Rauwolf, a German doctor and botanist who travelled widely in Asia and Africa in search of medicinal plants which had been mentioned by the early Arab and Greek physicians. Rauwolf published the results of his studies in 1582. Years later a new genus of the Apocynaceæ was named *Rauwolfia* in his honour. The *Rauwolfias* are found all over the world in tropical and sub-tropical regions and there are about 130 species. These will be found listed in the *Index Kewensis*[16,17]. Botanical and pharmacognostical descriptions will be found in Dymock[4] and other works[5,7,8,11–25].

Rauwolfia canescens, *R. sellowii*, *R. perakensis*, *R. micrantha*, *R. hirsuta* (*R. heterophylla*), *R. semperflorens*, *R. vomitoria* and *R. densiflora* are other *Rauwolfia* species which have aroused interest. Wan[26], Youngken[13,27], Datta and Mukherji[24], Trease and Evans[5], and Martinez[28], deal with *R. perakensis*[13,26], *R. canescens*[5,13], *R. micrantha*[13,27], *R. hirsuta*[28], and *R. densiflora*[13]. Youngken points out that the roots and leaves of *R. canescens* were also used in Ayurvedic medicine[13]. He discusses some of the adulterants of *R. serpentina*[13], (*R. canescens*, *R. densiflora*, *R. micrantha* and *R. perakensis*). *Ophiorrhiza mungos* and white- and red-flowered *Clerodendron* species have also been described[22,23].

Chemistry

This is reviewed in detail by Phillips and Chadha[29], and by others[8,30–34]. The chemistry of the *Rauwolfia* species will therefore be dealt with briefly. A large number of alkaloids have been isolated; of a number of these the structure has been determined and physical constants are described (Fig. 1, p. 472; Table I, p. 476). The clinically important alkaloids (reserpine and rescinnamine) differ chemically from the others.

Alkaloids from Rauwolfia serpentina *Benth.*

Reserpine. Although the root contains numerous other substances which in extracts, concentrates or in powdered whole root modify its action, reserpine is the main active principle of *Rauwolfia serpentina.*

Its isolation from an oleoresin fraction was reported by Muller, Schlittler and Bein[35]. Reserpine was obtained as a relatively weakly basic alkaloid with m.pt. 262–3° C. and $[\alpha]_D^{23}$ −117 to −118° in chloroform. In 1932, van Itallie and Steenhauer[36,37] had reported the isolation of three alkaloids A, B and C from *R. serpentina*. In a more recent publication[38] alkaloid B (m.pt. 262° C.) has been claimed by Steenhauer to be identical with reserpine. Muller and his colleagues[35] obtained reserpine from the oleoresin fraction[39–41]. The extraction of reserpine is also described by Dorfman and his colleagues[39] and by others[40,41] Reserpine is the 3:4:5-trimethoxybenzoic acid ester of reserpic acid[44–46]. In 1953 Klohs and his colleagues[42] assigned to it the empirical formula $C_{35}H_{44}O_{10}N_2$ and made a preliminary study of its structure. Furlenmeier and his colleagues[43] proposed the formula $C_{33}H_{40}O_9N_2$. The structure of reserpine has now been established[47–50] (Fig. 1, I) and it has been synthesised[50a]. More recently, the stereochemistry has been investigated[51–54]. Reserpine is an indole derivative and is related to yohimbine.

Other Alkaloids

With few exceptions[56] these are all indolic (see Table I). As pure substances they have so far little or no clinical importance but some have been studied pharmacologically.

Rescinnamine (Fig. 1, III). Like reserpine, this is a tertiary indolic base related to yohimbine. It was isolated from *R. serpentina* and characterised as the 3:4:5-trimethoxycinnamic acid ester of methyl reserpate in 1954–55 by Klohs and his colleagues[57,58] and also in 1954 by Haack and his colleagues[59]. This alkaloid was called reserpinine by Haack[59]. However, this name has been used to describe another rauwolfia alkaloid of different structure (*q.v.*) and there is a possibility of confusion arising.

Deserpidine[60,64]. This alkaloid is closely related to reserpine, lacking only the methoxy group of ring A[60–64] (Fig. 1, X). Although it has been reported to be present in *R. serpentina*, its main source is *R. canescens*[60–64]; it is also known as *canescine*[60–64] and *recanescine*[63].

Serpine. This tertiary indolic alkaloid has recently been isolated from an alcoholic extract of the roots of the Cochin variety of *R. serpentina* by Chatterjee and Bose[65]. It is related to the tetra-hydro-β-carbolines, yohimbine and rauwolscine. A structural formula has been proposed (Fig. 1, V).

Sarpagine. This is identical with raupine and with serpagine. Raupine was isolated from *R. serpentina* in 1953–54[66–68]. Sarpagine was also isolated from the same source in 1953[69]. In 1954 the identity of raupine with sarpagine was established[70]. The constitution of sarpagine has been studied by Raymond-Hamet[50] and Thomas[71]. It is a weakly basic tertiary indolic alkaloid related to yohimbine. The physical constants[69] are given in Table I, but according to Bodendorf and his colleagues[66] the molecular formula is $C_{20}H_{26}O_3N_2$ ($C_{19}H_{22}N_2O_2 + CH_3OH$), m.pt. 325°, $[\alpha]_D^{20} = +63°$ in acetic acid.

Rauhimbine and iso*Rauhimbine.* Rauhimbine has been shown to be identical with corynanthine[72], an alkaloid previously isolated from

Pseudocinchona africana Chev.[34,73,74]. *iso*Rauhimbine and rauhimbine were isolated from *R. serpentina* by Hofmann[75]. The molecular formula $C_{21}H_{26}O_3N_2$, together with chemical and physical data, showed both compounds to be isomers of yohimbine and to be closely related to rauwolscine, an alkaloid isolated from *R. canescens*. Studies on the constitution, etc., of *iso*rauhimbine have been made by Le Hir and his colleagues[76] and by Chatterjee and Talpatra[77]. A partial structural formula has been suggested[76].

Yohimbine. The well-known pharmacologically active indolic alkaloid yohimbine[34] has been isolated from *R. serpentina* by Bader and his colleagues[78], and by Hofmann[72].

Other Yohimbine Isomers. These have been isolated by Bader and his colleagues[79] (alkaloid 3078); Weisenborn and his colleagues[80] (δ-yohimbine) (Fig. 1, VI); Hofmann[56] (δ-yohimbine, alkaloid C = an 11-methoxy-δ-yohimbine); Bader and his colleagues[81] (3-*epi*-α-yohimbine = 3-*epi*rauwolscine = alkaloid 3078)[79].

Reserpinine. This is another tertiary indolic base and is related to the alkalid tetrahydroalstonine. Reserpinine was isolated by Schlittler and his colleagues[82] in 1954. The m.pt. of the isolated compound was 238–9° C. (corr.) and a structural formula was proposed (Fig. 1, IV).' Weisenborn and his colleagues[80] also isolated reserpinine. The "alkaloid A" of Neuss and his colleagues[83] with the molecular formula $C_{22}H_{26}N_2O_4$ and related to tetrahydroalstonine is the "substance I" of Popelak and his colleagues[67], and is identical with the reserpinine of Schlittler and Weisenborn and their colleagues[80,82]. Recently, Janot and Le Men[84] have shown that *Vinca major* L. contains reserpinine. Raubasinine[59,67,85] is identical with reserpinine. Hofmann's "alkaloid C"[72] may also be reserpinine.

Reserpiline. Reserpiline[86] is closely related to reserpinine. It is an amorphous alkaloid possessing an additional methoxyl group (Fig. 1, XI).

Ajmaline (Fig. 1, IX). Ajmaline is from the chemist's point of view perhaps the most interesting and most studied of the many *Rauwolfia* alkaloids. It is a tertiary indoline base. Much work has been done upon the elucidation of its structure which is not settled. Robinson[97] has suggested a possible structure, referred to in some detail by Phillips and Chadha[29]. Ajmaline was first isolated in 1931 by S. and R. H. Siddiqui[87] who, in addition, obtained in crystalline condition the alkaloids ajmalinine, ajmalicine, serpentine and serpentinine. The molecular formula $C_{21}H_{26}O_2N_2$ was proposed[87] for ajmaline. Van Itallie and Steenhauer[37] isolated ajmaline at about the same time. Studies on the structure of ajmaline have been carried out by the Siddiquis[87–90] and others[91–97]. Notable amongst these has been the work of Robinson and his colleagues at Oxford[92,93,96,97]. It is considered identical with rauwolfine[29,36].

iso*Ajmaline*. This was isolated by Siddiqui[90] from a variety of *R. serpentina* known as "Dehra Dun". It is associated with *neo*ajmaline. Siddiqui[90] pointed out that the alkaloid content of "Dehra Dun" *R. serpentina* differed in yield and in character from that of the "Bihar" variety.

*iso*Ajmaline is formed when ajmaline or *neo*ajmaline are heated. *neo*-Ajmaline on heating melts at 205°; if heating is continued the melt re-solidifies forming *iso*ajmaline which melts at 264–266°. *iso*Ajmaline is an isomer of ajmaline[93,96].

Rauwolfinine[98–102]. This is an indoline alkaloid. Its structure is not completely known.

Serpentine[80,88,89,103–105]. This is a yellow quaternary indolic, anhydronium base. Siddiqui and Siddiqui suggested that its molecular formula was $C_{20}H_{20}O_3N_2 \cdot 1\frac{1}{2}\ H_2O$[87–89]. This has now been revised to $C_{21}H_{22}O_3N_2$[103]. It is a stereoisomer of alstonine[80] and very closely related to the alkaloid rauwolscine which occurs in *R. canescens*[106–111]. A structural formula has been proposed by Klohs and his colleagues[105] (Fig. 1, VII).

Serpentinine. Serpentinine isolated by the Siddiquis[87–89] and later by Schlittler and his colleagues[112,113] appears to be an indolic anhydronium base[112,113].

Ajmalicine. This is known also as π-tetrahydro serpentine. It was isolated from *R. serpentina* by Klohs and his colleagues[105], who proposed a structure for it (Fig. 1, VI). It is closely related to serpentine (Fig. 1, VII).

Minor alkaloids. The following appear in the literature: ajmalinine[87–89], serpinine[114], chandrine[115], methyl reserpate[56], R.S.51[116], thebaine[56], papaverine[56].

Alkaloids from other Rauwolfia *Species*

Rauwolfia canescens Linn. *R. canescens* contains reserpine[61,64,117], rauwolscine (α-yohimbine)[61,107,108,109,110,121,122], yohimbine[61], serpentine[61] canescine[61,62], pseudoyohimbine[61], recanescine[63] (probably the same as deserpidine[61,64] and 11-desmethoxyreserpine). Canescine and recanescine are probably identical[63]. β-Yohimbine has been isolated by Hofmann[118] from *R. canescens* roots. Stoll and his colleagues have isolated aricin, *iso*reserpinine, reserpiline and *iso*reserpiline from *R. canescens* leaves[119]. The alkaloids of *R. canescens* have at the moment more chemical than pharmacological interest. They are indolic and have a close relation to the *R. serpentina* alkaloids. The isolation of aricin (Fig. 1, VIII) is of interest. Aricin is a 10-methoxy derivative of ajmalicine; it is isomeric with reserpinine. Stoll and his colleagues[119] point out that there are biogenetic similarities between the rauwolfia and strychnos alkaloids.

Rauwolfia hirsuta Jacq. (= *R. heterophylla* Roem and Schult.) This species grows in South and Central America and in Mexico. Martinez[28] describes its occurrence in Mexico. Guatemalan *R. hirsuta* is known as "chalchupa"[123], the Colombian plant as "pinique-pinique"[124,125]. The following substances have been isolated from *R. hirsuta*: the chalchupins A and B[123], reserpine[126–129] and narcotine[126], rauwolscine[127,129] and alstonine[127,131] (Fig. 1, VII), serpentine[128–130], ajmaline[128], ajmalicine[129] yohimbine[129], heterophyllin (aricin = 5-methoxyajmalicine)[129], alkaloid A[131], rautensin (total alkaloid fraction)[131].

Rauwolfia micrantha Hook. Rao and Rao[132] have reported the isolation of bases A, B and C from an oleoresin fraction. Youngken[27] describes the microscopy of the roots and tests for alkaloids. It is also known as Malabar rauwolfia.

Rauwolfia sellowii. The alkaloid content of the various plant organs has been determined[133]. Seba and his colleagues indicate that *R. sellowii* contains ajmaline and serpentine[134].

Quantitative Estimation of Rauwolfia Alkaloids

Rauwolfia extracts, powdered root and reserpine are being widely used. They are available in various forms, including tablets, mixtures and solutions for injection. The reserpine content or hypotensive potency may be determined in a number of ways: Bakshi[135] pointed out that the *R. serpentina* alkaloids gave a blue fluorescence in ultra-violet light. This property was made use of in an assay method. Sheppard and his colleagues[136] exposed chloroform solutions of reserpine to a given intensity of ultra-violet light for a given period of time and measured the resulting fluorescence. They showed that the relationship concentration/fluorescence was linear over a range of 0·1 to 1·0 μg. of reserpine per ml.

McMullen and his colleagues[137] have studied the physical and chemical properties of the *R. serpentina* alkaloids with the objects of identifying and estimating pure reserpine, and reserpine in pharmaceutical preparations containing either the pure base or a mixture of alkaloids. They noted that the m.pt. was a good criterion of purity of reserpine, that the infra-red spectrum was an excellent method of identification and that the ultra-violet spectrum could, suitably modified, be used as a quantitative method of analysis. Three assay methods are described, together with a chromatographic method for separating reserpine from other rauwolfia alkaloids. Banes[138] pointed out that the ultra-violet absorption spectrum was not satisfactory for use in an assay procedure unless the reserpine had been isolated. In the absence of recanescine, identification of trimethoxybenzoic acid amongst the products after saponification of an alkaloid mixture indicated the presence of reserpine. Reserpine may be determined by use of a suitable method of extraction, treatment with vanillin and determination of the absorption at 532 mμ. In strongly acidic solution reserpine and reserpic acid give a red colour with vanillin. Another method of assay is described by Booth[139]. This is suitable for elixirs and depends upon the formation of a chloroform-soluble reserpine-bromophenol blue complex. The absorbance of the solution at 402 mμ is read. Sakal and Merrill[140] describe a spectrophotometric method for reserpine. Horhammer and his colleagues[141,142] describe methods suitable for extraction and determination of *R. serpentina* alkaloids from the plant.

In a drug so widely used and with such interesting properties as reserpine, it is inevitable that methods should be sought for its estimation in tissue and in body fluids, etc. These methods are generally based upon the fluorimetric, colorimetric and spectrophotometric characteristics. Fluorimetric and spectrophotometric methods are being used by Gillis and Lewis[143] to attempt to follow the distribution of reserpine in the rat's brain.

Fluorimetric methods for its estimation in blood and urine are described by Kelly[144] and others[145,146]. Sheppard and his colleagues[147] have labelled reserpine with ^{14}C in the 4-methoxy group of the trimethoxybenzoic acid moiety and have administered this compound to adult male rats with the object of studying the distribution of ^{14}C. The significance of their results will be discussed later.

PHARMACOLOGY

Although many careful studies have been made of the pharmacology of reserpine, it would be hazardous to assign to it a precise site or mode of action. In addition, pharmacological studies have been made upon extracts and concentrates of the whole root of *R. serpentina* and upon the now numerous purified alkaloids. Reserpine is without doubt the most studied and most important of these, and will be dealt with in some detail. The continued use and importance of extracts and of the powdered whole root makes it impossible to overlook the other alkaloids and preparations.

In 1931, Sen and Bose[148] observed that the dried root of *R. serpentina* contained 1 per cent. of total alkaloids which caused a slight fall in blood pressure with stimulation of respiration and relaxation of smooth muscle in the cat. They also observed a lowering of the blood pressure and sedation when the powdered drug was given to patients. In the same year Roy[149] noted that large doses of *R. serpentina* caused sleep, dulling of sensations, diminished reflexes and, if the dose was lethal, death from respiratory failure, the heart continuing to beat for some time. The work of Chopra and his colleagues[1] which followed the earlier investigations was both interesting and important. The alkaloid investigated was a dull brown-yellow substance and was used in 1 per cent. solution. It was toxic to *Paramœcium caudatum* and to white mice, cats and guinea-pigs. In chloralosed cats increased intestinal tonus and peristalsis was observed. Five to 10 mg. slowed the rate and decreased the depth of respiration. There was a fall in blood pressure partly due to cardiac slowing and partly to vasodilatation. There was stimulation of the virgin or pregnant cat uterus *in situ*. Chopra also noted sedation, hypnosis, impairment of sensory perceptions and of reflexes in frogs, and drowsiness and quietning in other species. The alkaloid of Chopra and his colleagues[1] was thought to be ajmaline, and was shown in 1940 by Raymond-Hamet[150,151] to be a sympatholytic.

Other work on the rauwolfia alkaloids followed from Calcutta[8,152–154]. The "Bihar" variety of *R. serpentina* was shown to be more hypnotic than that from Dehra Dun[152]. The pharmacology of serpentine, serpentinine and ajmaline were also studied. These acted as convulsant poisons in rats. Alcoholic extracts of *R. serpentina* were depressant. The total alkaloid hydrochlorides had a hypnotic effect. Alcoholic extracts and the total alkaloids antagonised picrotoxin, however the actions of ajmaline, serpentine and serpentinine were summated with those of the analeptic[152]. The total alkaloids and alcoholic extract were hypotensive, serpentine being

thought the chief hypotensive agent while ajmaline and serpentinine caused hypertension[153,154].

Studies were also made of the comparative sedative and hypotensive potencies of extracts and the purified total alkaloids from Bengal, Bihar

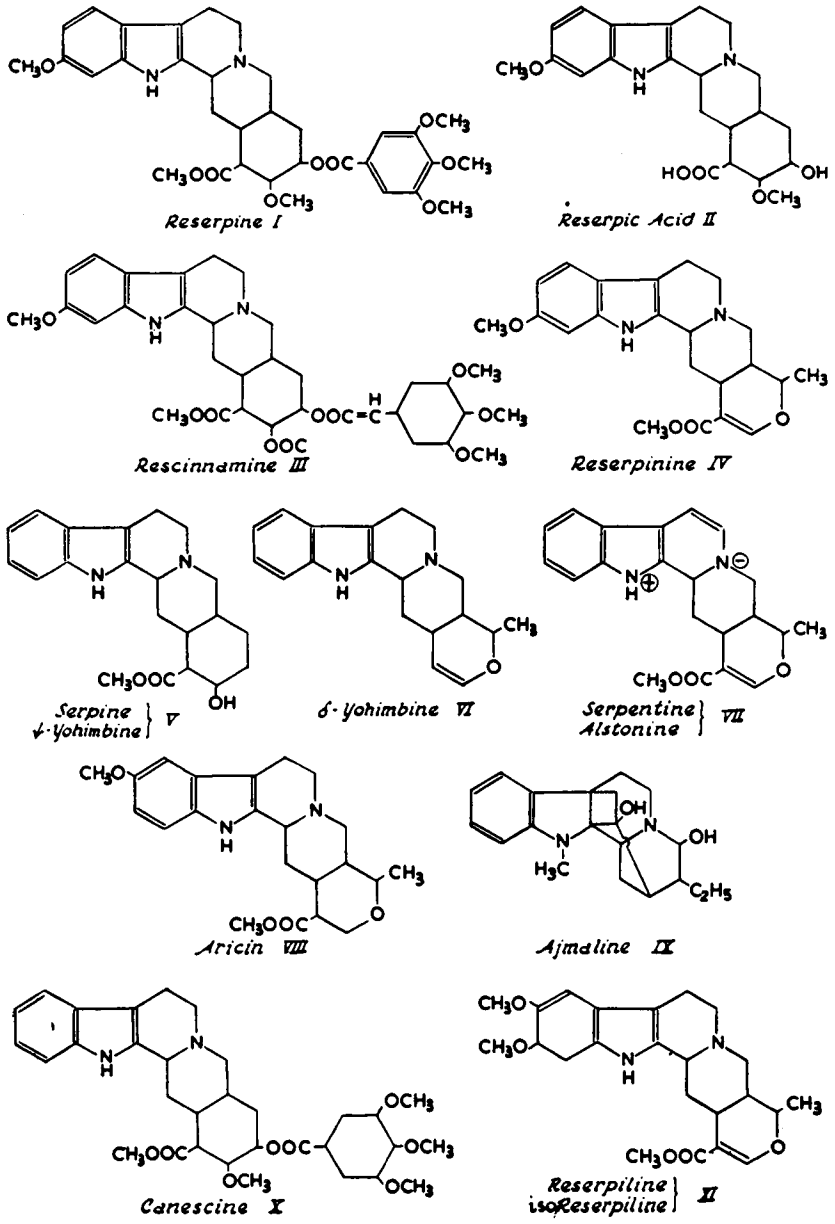

FIG. 1. The structure of some of the rauwolfia alkaloids.

and Dehra Dun varieties of *R. serpentina*[155–157]. An alcohol soluble, oil-free resin, free from alkaloids, was isolated. This caused sedation and hypnosis which were delayed in onset for 3 to 4 hours. The resin was synergistic with ether and chloralose and antagonistic to picrotoxin. Its site of action was thought to be the hypothalamus[155,156]. Gupta and his colleagues[158] investigated the hypnotic resin fraction further. It is interesting to note that Dymock[159] in 1891 isolated a crude resin fraction from *R. serpentina* (the root was shown to contain alkaloids at about the same time)[160,161]. Gupta and Kahali[157] noted that the *R. serpentina* alkaloids appeared to depress the vasomotor centre and also to act peripherally.

Before dealing with the pharmacology of reserpine, some of the later studies made upon extracts and concentrates and the whole root of *R. serpentina* will be considered.

Arnold[162] considered that *R. serpentina* acted as a sympatholytic agent with a marked central effect. Studies on dogs were made by Kramer and his colleagues[163] which showed that extracts lowered blood pressure and counteracted the effects of sympathomimetics.

A number of studies were made upon the actions of *R. serpentina* on vasomotor reflexes. Werner and his colleagues[164] elicited the carotid sinus reflex in cats by temporary occlusion of the common carotid arteries, and then, by artificially raising the intra-carotid pressure, produced a depressor response. Crude extracts of the roots of "Dehra Dun" *R. serpentina* reduced both pressor and depressor responses. Antagonism to the pressor effects of 100 m. units of pitressin was not shown, nor were cyanide-elicited reflexes antagonised. Werner and his colleagues[164–169] observed a non-competitive peripheral adrenergic blocking activity not primarily related to the vasomotor reflex blocking action. Work with the total purified alkaloids and with some of the purified bases, in which these were given intra-cisternally, pointed to a central action being responsible for the lowering of blood pressure and the blockade of vasomotor reflexes. Intracranial application of the *R. serpentina* alkaloids in animals decerebrated at the mid-collicular level did not influence the mean arterial blood pressure, nor did it abolish the pressor response to baroceptor stimulation. The extracts were assumed to interfere with the central control of vasomotor reflex activity and tone.

Vasodilatation in the kidney after injection of *R. serpentina* has also been observed in chloralosed cats. An alcoholic extract of *R. serpentina* (Dehra Dun) roots produced marked vasodilatation and blocked hæmorrhage-induced vasoconstriction in the renal vascular bed[169].

Other workers have investigated alkaloidal fractions from *R. serpentina*. Rubin and Burke[170] gave whole root powder to trained dogs for periods of 2 weeks or longer. The animals showed hypotension, bradycardia, sedation, miosis, relaxation of the nictitating membrane, ptosis, diarrhœa and tremor at dose levels of 80 and 320 mg. per kg. per day, the symptoms appearing within 48 hours. Bradycardia and miosis were blocked by atropine. At higher dose levels (320 mg. per kg. per day) there was

ultimately anorexia with emaciation and dehydration followed by death. Using a standardised alkaloidal extract of *R. serpentina* Gourzis and his colleagues[171] showed that hypotensive doses caused vasodepression and bradycardia. There was an increased pressor response to adrenaline and enhancement of the hypotensive and cardio-accelerator actions of isoprenaline. The carotid sinus reflex was reduced and the rise in blood pressure due to hypoxia was abolished. No change was seen in the responses to acetylcholine, histamine and efferent vagal stimulation but the hypertensive response which follows electrical stimulation of the afferent vagus was blocked.

Lim and his colleagues[173] have recently compared the responses to injections into the fourth cerebral ventricle and the carotid artery with intravenous administration of rauwolfia alkaloids, veratrum alkaloids and hydrallazine. Rauwolfia is considered to cause hypotension primarily by a central effect. Studies on the circulatory effects of *R. serpentina* have also been made by other workers[174,175]. Thuillier and Mouille[176] have shown that an extract of *R. serpentina* inhibits the action of acetylcholine on the isolated guinea-pig intestine. Kronheim and Koster[177] have noted a transient fall in adrenal ascorbic acid concentration after administration of either an alkaloid extract of *R. serpentina*, or of reserpine, serpentine, ajmaline or alkaloid F. A mixed alkaloid preparation of *R. serpentina* had no effect on the basal metabolic rate in rats[178].

Cronheim and Toekes[180] investigated the sedative actions of an alkaloidal extract of *R. serpentina*. Sedation was observed in dogs, cats, guinea-pigs and mice together with antagonism to the central excitant action of amphetamine in mice. Achelis and Kroneberg[181] compared the effects of the total alkaloids with those of reserpine on the dog's blood pressure. Hypotension due to the total alkaloids was not thought to result from the small amount of reserpine present since it was of speedy onset and long duration. Hypotension induced by reserpine on the other hand shows a slow onset coupled with bradycardia.

Gourzis[182,183] has shown that sedative doses of an alkaloidal extract of *R. serpentina*, unlike phenobarbitone, raise the emetic threshold to vomiting in veratrum-preparation treated dogs. A mouse ptosis bioassay of *R. serpentina* for reserpine-like activity has been described by Rubin and Burke[184].

Working mainly upon isolated tissue preparations, Banerjee and Lewis[185] showed that the alseroxylon fraction of the alkaloids of *R. serpentina* appeared to possess anticholinergic properties on skeletal, smooth and cardiac muscle. The effects were persistent and in some tissues there was a latent period before the maximum effect was shown. A delayed fall in body temperature in mice was observed.

The Pharmacology of Reserpine

In 1952 Muller, Schlittler and Bein[35] reported the isolation of reserpine —a weak base with a prolonged sedative action—from the oleoresin fraction of an extract of *R. serpentina*. The typical sedative effects of *R. serpentina* extracts were due after all to an alkaloid.

In 1953 Bein[186] showed that reserpine had hypnotic and hypotensive actions; it inhibited the pressor responses to electrical stimulation of the afferent vagi and sciatic nerves and also the carotid sinus pressor reflex. Pressor effects arising in response to electrical stimulation of the splanchnic nerves were not inhibited. The drug possessed no peripheral adrenergic- or ganglion-blocking activity. It potentiated the pressor action of adrenaline, noradrenaline and ephedrine. The hypotensive effect was not inhibited by atropinisation or vagotomy and there was no neuromuscular blocking activity. Peristalsis was stimulated. Bein considered that reserpine acted on the central nervous system. In the same year Bein and his colleagues[187,188] pointed out that reserpine not only caused sedation, hypnosis and hypotension, but was capable of inducing bradycardia and miosis, and that it also had laxative and body temperature-lowering effects. The Swiss investigators noted that reserpine was not a narcotic, and differed from the barbiturates and bromides. Reserpine-treated animals appeared tired and dazed, and slept peacefully but could always be wakened. It was felt that these long-lasting and delayed actions might be due to insolubility of the alkaloid or to its action being due to the need for prior formation of an active breakdown product. No direct peripheral vasodilator action was observed. Reserpine's action was not comparable with that of veratrine. A direct effect upon the sympathetic autonomic centres in the brain regulating the blood pressure was postulated and attention was drawn to the resemblance between the pharmacological effects of reserpine and the effects produced in 1947 by Hess[189] after stimulation of the mesencephalon. Hess's trophotropic-endophylactic system on stimulation gives rise to adynamia or sleep, miosis, relaxation of the nictitating membrane, hypotension, bradycardia, inhibition of respiration, hypothermia and defæcation. The analogy with reserpine is obvious.

During 1953 a number of contributions to the study of reserpine were made in America. Trapold and others[190,191] showed that reserpine in dogs caused respiratory depression, increased intestinal mobility, miosis, persistent hypotension and bradycardia, the miosis and bradycardia being abolished by atropine. One mg. per kg. of reserpine did not significantly depress cardiac stroke volume, output and index, as measured by Fick's method, until maximum hypotension was established. Reserpine produced vasodilatation which was the initial cause of its hypotensive effects. Maintenance of hypotension was due to reduced cardiac output, but cardiovascular changes seemed to be secondary to depression of the central nervous system, the drug acting on higher centres such as those of the hypothalamus. Plummer and his colleagues[192] confirmed the effects of reserpine in the dog, rat and rabbit. Cronheim and his colleagues[193] pointed out that reserpine was the most potent single *Rauwolfia serpentina* alkaloid so far examined, and noted that other active alkaloids present (ajmaline, ajmalicine, serpentine, serpentinine and alkaloid G) had not shown hypotensive activity in doses of 1 to 3 mg./kg./day in dogs.

Further contributions came from Swiss workers in 1954[194–199]. Again, hypotension and sedation of long duration were noted. Sedation was

(continued on p. 480.)

TABLE I THE RAUWOLFIA ALKALOIDS

R.s. = *Rauwolfia serpentina* Benth. *R.h.* = *R. hirsuta* Jacq. *R.sl.* = *R. sellowii.*
R.c. = *R. canescens* L. *R.m.* = *R. micrantha* Hook. *R.sm.* = *R. semperflorens* Schlecht.
R.t. = *R. tetraphylla* L. *R.v.* = *R. vomitoria* Afz. *R.o.* = *R. obscura.*
R.cf. = *R. caffra* (Sonder)

Alkaloid	Plant source	Identical with	Molecular formula	Physical constants			References
				m.pt. °C.	$[\alpha]_D$	$[\alpha]_{5461}$	
Ajmalicine	*R.s. R.h.*	Alkaloid F π-Tetrahydro serpentine δ-Yohimbine	$C_{21}H_{24}O_3N_2$	250–2	$[\alpha]_D^{23}$ − 48·5° (C_5H_5N)	—	105
Ajmaline	*R.s. R.h. R.sl.*	Rauwolfine	$C_{20}H_{26}O_2N_2$	205	$[\alpha]_D^{33}$ + 128° ($CHCl_3$)	—	29, 97, 87–97
Ajmalinine	*R.s.*	*Alkaloid C	$C_{20}H_{26}O_3N_2$	180–1	$[\alpha]_D^{33}$ − 97° ($CHCl_3$)	—	87–89
Alstonine	*R.v.* *R.o.*	—	$C_{21}H_{24}O_3N_2$	225–6	*R.v.* $[\alpha]_D^{20}$ − 115 ± 4° ($CHCl_3$) *R.o.* $[\alpha]_D^{22}$ − 109 ± 6° ($CHCl_3$)	—	127, 131
Aricin	*R.c. R.h.*		$C_{22}H_{26}O_4N_2$	190		—	129
Alkaloid A	*R.s.*	Reserpinine alkaloid C	$C_{22}H_{26}O_4N_2$	240–1		—	131
*Alkaloid C	*R.s.*	Ajmalinine		177	$[\alpha]_D$ = − 76·4°	—	87–89
Alkaloid C	*R.s.*	Reserpinine, alkaloid A	$C_{22}H_{26}O_4N_2$	240–1	$[\alpha]_D^{20}$ = − 127° (C_5H_5N)	—	56
Alkaloid F	*R.s.*	Ajmalicine π-Tetrahydro serpentine	$C_{21}H_{24}O_3N_2$	253–4	$[\alpha]_D^{20}$ − 37 ± 6° (CH_3OH)	—	105
Alkaloid 3078	*R.s.*	3-*epi*-α-Yohimbine 3-*epi*Rauwolscine	$C_{21}H_{26}O_3N_2$	125–8 181–3	$[\alpha]_D^{26}$ − 96° (C_5H_5N)	—	79
Base A	*R.m.*	—	—	264–6	—	—	132
Base B..	*R.m.*	—	—	247–8	—	—	132
Base C	*R.m.*	—	—	157–9	—	—	132

TABLE I—*continued*

Alkaloid	Plant source	Identical with	Molecular formula	Physical constants: m.pt. °C.	Physical constants: $[\alpha]_D$	Physical constants: $[\alpha]_{5461}$	References
Canescine	*R.c.*	11-Desmethoxy reserpine	$C_{32}H_{38}O_9N_2$	230–4	$[\alpha]_D - 163 \pm 2°$	—	60–64
Chalchupine A	*R.h.*	Rauwolscine	—	230–2	—	—	123
Chalchupine B	*R.h.*	—	—	—	—	—	123
Chandrine	*R.s.*	—	$C_{21}H_{26}O_3N_2$	230–1	—	—	115
Corynanthine	*R.s.*	Rauhimbine	$C_{21}H_{26}O_3N_2$	218–25	$[\alpha]_D^{20} - 82°$ (C_5H_5N)	—	34, 72–74, 75
Deserpidine	*R.c.*	Recanescine, 11-Desmethoxy reserpine	$C_{32}H_{37}O_8N_2$	228–32	$[\alpha]_D^{24.5} + 137°$ ($CHCl_3$)	—	60–64
11-Desmethoxy reserpine	*R.c.*	Deserpidine Recanescine	—	228–32	$[\alpha]_D^{24.5} + 137°$ [$CHCl_3$]	—	60–64
Heterophyllin	*R.h.*	Aricin	$C_{22}H_{26}O_4N_2$	190	—	—	129,119
*iso*Ajmaline	*R.s.*	—	$C_{20}H_{26}O_2N_2$	264–6	$[\alpha]_D^{35} + 72{\cdot}8°$ (C_2H_5OH)	—	90, 93, 96
*iso*Reserpiline	*R.c.*	—	$C_{32}H_{38}O_9N_2$	211–2	—	—	119
*iso*Reserpinine	*R.c.*	—	$C_{32}H_{38}O_9N_2$	225–6	—	—	119
*iso*Rauhimbine	*R.s.*	—	$C_{21}H_{26}O_3N_2$	225–7	$[\alpha]_D^{20} - 104°$ (C_5H_5N)	$[\alpha]_{5461}^{20} - 129°$ (C_5H_5N)	75, 76, 77
Methyl reserpate	*R.s.*	—	$C_{23}H_{30}O_5N_2$	244–5	$[\alpha]_D^{20} - 106°$ (C_5H_5N)	—	56
Narcotine	*R.h.*	—	$C_{20}H_{23}O_7N$	205–7	—	—	126
*neo*Ajmaline	*R.s.*	—	$C_{20}H_{26}O_2N_2$	205–7	—	—	90, 250
Papaverine	*R.s.*	—	$C_{20}H_{21}O_4N$	147	—	—	56
Pseudoyohimbine	*R.c.*	—	—	265–78	—	—	61
Raubasine	*R.s.*	Substance II δ-Yohimbine Alkaloid F Ajmalicine π-Tetrahydro-serpentine	—	—	—	—	—

TABLE I—*continued*

Alkaloid	Plant source	Identical with	Molecular formula	Physical constants			References
				m.pt. °C.	$[\alpha]_D$	$[\alpha]_{5461}$	
Raubasinine	*R.s.*	Substance I Alkaloid C Reserpinine Alkaloid A	—	—	—	—	—
Rauhimbine	*R.s.*	—	$C_{21}H_{26}O_3N_2$	218–25	$[\alpha]_D^{20}$ − 82° (C_5H_5N)	$[\alpha]_{5461}^{20}$ − 94° (C_5H_5N)	72, 34, 73, 74, 75
Raupine	*R.s.*	Sarpagine + CH_3OH	$C_{20}H_{24}O_3N_2$	325	$[\alpha]_D^{20}$ + 63° (CH_3COOH)	—	50, 66–71
Rauwolfine	*R.s. R.h.*	Ajmaline	$C_{20}H_{24}O_3N_2$	205	$[\alpha]_D^{33}$ + 128° ($CHCl_3$)	—	29, 97, 87–97
Rauwolfinine	*R.s.*	—	$C_{19}H_{26}O_3N_2$	235–6	$[\alpha]_D^{32}$ − 34·7°	—	98–102
Rauwolscine	*R.h. R.c.*	α-Yohimbine	$C_{21}H_{26}O_3N_2$	—	—	—	61, 107–110, 121, 122
Rauwolfine	*R.cf. R.s.*	Ajmaline	$C_{21}H_{26}O_2N_2$	160	—	—	274
Recanescine	*R.c.*	11-Desmethoxy reserpine Deserpidine	$C_{22}H_{27}O_8N_2$	—	—	—	60–64
Rescinnamine	*R.s.*	—	$C_{35}H_{42}O_9N_2$	238–9	$[\alpha]_D^{24}$ − 97° ($CHCl_3$)	—	57–59
Reserpiline	*R.s. R.c.*	—	$C_{33}H_{38}O_6N_2$	—	$[\alpha]_D^{24}$ − 40° (C_2H_5OH)	—	86, 119
Reserpine	*R.s. R.c. R.h.*	—	$C_{33}H_{40}O_9N_2$	262–6 277–8 (corr.)	$[\alpha]_D^{23}$ − 117·8° ($CHCl_3$)	—	35–46, 51–54
Reserpinine	*R.c. R.s.*	Alkaloid A	$C_{33}H_{36}O_4N_2$	240–1	$[\alpha]_D^{23}$ − 117° ($CHCl_3$)	—	80, 82, 83, 67, 84
Sarpagine (Serpagine) ..	*R.s. R.h.*	Raupine − CH_3OH	$C_{19}H_{22}O_3N_2$	320	$[\alpha]_D^{20}$ + 54° (C_5H_5N)	—	50, 66–69, 71
Serpenine	*R.s.*	—	—	315	—	—	114
Serpentine	*R.h.* *R.c. R.sl.* *R.s.*	—	$C_{21}H_{20}O_4N_2$	157–8	$[\alpha]_D^{40}$ + 188° (H_2O)	—	106–111, 80, 87–89, 103–105

TABLE I—*continued*

Alkaloid	Plant source	Identical with	Molecular formula	Physical constants			References
				m.pt. °C.	$[\alpha]_D$	$[\alpha]_{5461}$	
Serpentinine	*R.s.*	—	$C_{20}H_{20}O_3N_2$	263–5	—	—	87–89, 112, 113
Serpine	*R.s.*	—	$C_{21}H_{26}O_3N_2$	213	$[\alpha]_D^{20}$ + 70·1° (C_5H_5N)	—	65
Substance I	*R.s.*	Alkaloid A Reserpinine	$C_{22}H_{26}O_4N_2$	228	$[\alpha]_D^{20}$ − 123° ($CHCl_3$)	—	—
Substance II	*R.s.*	Raubasine Alkaloid F Ajmalicine π-Tetrahydro-serpentine δ-Yohimbine	$C_{21}H_{24}O_3N_2$	247–8	$[\alpha]_D^{20}$ − 61° ($CHCl_3$)	—	—
π-Tetrahydroserpentine ..	*R.h. R.s.*	Ajmalicine	$C_{21}H_{24}O_3N_2$	250–2	$[\alpha]_D^{23}$ − 48·5° (C_5H_5N)	—	105
Tetraphyllicine	*R.t.*	—	$C_{20}H_{20}N_2$	320–2	$[\alpha]_D^{27}$ + 21° (C_5H_5N)	—	—
Tetraphyllin	*R.t.*	—	$C_{22}H_{26}O_4N_2$	220–3	$[\alpha]_D^{28}$ − 73°($CHCl_3$) − 35°(C_5H_5N)		
Thebaine	*R.s.*	—	$C_{19}H_{21}O_2N$	195	$[\alpha]_D^{20}$ − 279° (C_5H_5N)	—	56
R.S.51	*R.s.*	—	—	—	—	—	116
3-*epi*-α-Yohimbine	*R.s.*	Alkaloid 3078 3-*epi*-Rauwolscine	$C_{21}H_{26}O_3N_2$	125–8 181–3	$[\alpha]_D^{26}$ − 96° (C_5H_5N)	—	79
α-Yohimbine	*R.c.*	Rauwolscine	—	—	—	—	—
δ-Yohimbine	*R.h. R.s.*	Alkaloid F Ajmalicine π-Tetrahydro sepentine	$C_{21}H_{24}O_3N_2$	258–9	$[\alpha]_D^{20}$ − 45° (C_5H_5N)	—	80
β-Yohimbine	*R.c.*	—	$C_{21}H_{26}O_3N$	246–9	—	—	—
Yohimbine	*R.c. R.h. R.s.*	—	$C_{21}H_{26}O_3N_2$	235–7	$[\alpha]_D^{20}$ + 105° (C_5H_5N)	—	34, 72, 78

different from that produced by phenobarbitone or sodium bromide as shown by EEG studies. Reserpine 10^{-6} was shown to inhibit barium-chloride constriction of the isolated blood vessels of the rabbit's leg, but not to cause dilatation by itself. There was dilatation of the coronary vessels of the mammalian heart which was inhibited by tripelennamine, phentolamine, barium chloride and acetylcholine but not by atropine. Reserpine was shown to inhibit the constriction caused by barium chloride and pitressin but it does not antagonise the actions of histamine, adrenaline or noradrenaline. Similar results were obtained by Gillis and Lewis[143]. Reserpine antagonised the characteristic central effects of caffeine, cocaine, morphine and hyoscine, but not convulsions due to leptazol, nicotine or picrotoxin. In experiments on isolated organs[197] reserpine was used in a solution with ascorbic acid or it was dissolved in a mixture of propylene glycol, ethanol and distilled water. These solubilising agents may modify the drug's effects, and need careful control experiments[197].

A number of contributions from America appeared in 1954. Schneider and Earl[200] compared the sedative effects induced by reserpine and barbiturates in monkeys and showed that these differed from one another in the influence upon the EEG and behaviour. Plummer and his colleagues,[201], discussing their own and other investigators' findings, pointed out that reserpine induced quiet and sedation in monkey, dog, cat, rabbit, guinea-pig, rat and mouse. Dogs and guinea-pigs were very susceptible, the monkey much less so. The latency and prolongation of sedation was seen, but there was no tolerance to the drug. Reserpine was primarily a tranquilising and inactivating agent allowing but not inducing sleep. Augmentation of secretory and motor activity of the dog gastrointestinal tract was seen, with increase in volume and hydrochloric acid content of the gastric juice. Oxyphenonium (an anticholinergic agent) eliminated the increased secretion.

Plummer and his colleagues[201] noted that the acute toxicity of reserpine is low—monkeys tolerate 400 mg. per kg. per day by mouth, and rats 1 g. per kg. per day by mouth. There is profound sedation but the animals always recover. Monkeys tolerate 4 mg. per kg. intravenously and rats 8 mg. per kg. intravenously, larger doses cannot be given owing to the insolubility of the drug. The dosage in dogs was limited because of the complication of diarrhœa. Rats, dogs and monkeys have been given as much as 4, 35 and 3 mg. per kg. per day respectively by mouth for as long as six months without sign of undue toxicity.

Reserpine hypotension was thought[191,202] to be initiated by central nervous system depression, possibly hypothalamic in location, which caused vascular relaxation followed by a reduction in cardiac output. In dogs a peripheral action was not demonstrable[191,202].

Barrett and his colleagues[203] studied the actions of reserpine on gastric motility in the guinea-pig, cat, dog and rabbit. No peripheral cholinergic activity was found using the isolated ileum and colon. Anticholinergic activity was shown on the acetylcholine-stimulated ileum of guinea-pig, dog and rabbit but little effect was seen when cat tissues were used. There

was slight antagonism to acetylcholine activity on the isolated rabbit colon but no influence on the colon of the dog or cat. In the dog, reserpine caused gastric secretion inhibited by oxyphenonium but not by tripelennamine. Esterification was shown to be necessary for reserpine-activity by Plummer and his colleagues[204]. Reserpic acid and trimethoxy-benzoic acid both lacked sedative and hypnotic properties[204]. Schneider[205] showed that reserpine antagonised the effects of morphine in white mice subjected to a beam of heat focussed on the tip of the tail but itself did not change the pain threshold. Rauwolfia extracts and reserpine were found by Jenney[206] to lower the electroshock seizure threshold significantly in mice.

Chen and his colleagues[207,208] showed that reserpine exerted a facilitation reaction on the central nervous system in mice and potentiated the convulsive effects of leptazol and caffeine but not of strychnine. There was reduction of the electrically-induced convulsive seizure threshold. The anticonvulsant actions of phenytoin and other central nervous system depressants were opposed by reserpine. Moyer and others (1954)[209] studied changes in cardiovascular and renal hæmodynamics after reserpine administration in dogs. Renal hæmodynamics and electrolyte secretion were not significantly altered and no consistent effect could be observed on cardiac output.

A number of recent publications deal with the actions of reserpine on the nervous system. Longo and Napolitano[212] examined the effects of reserpine administration on the rabbit EEG and on the response to hypothalamic stimulation in the rabbit. The first phase after drug administration was one of excitation with decreased rest periods, prolongation of active periods and occasional outbursts of spike forms. This phase was followed by a blockade of the response usually elicited by sensory perceptions, and the motor and emotional responses caused by electrical stimulation of the hypothalamus were much reduced or abolished. Reserpine was thought to act on the diencephalic nuclei by depressing the ascending and descending pathways of the reticular formation. Gangloff and Monnier[213] stimulated the cortex, diencephalon, rhinencephalon and reticular formation of the rabbit's brain, and recorded and studied spontaneous brain activity and the electrically induced discharge in the cortex, the dorsal, medial and lateral thalamic nuclei, the rhinencephalon and the reticular formation of the brain stem. Reserpine was shown to depress the diencephalo-cortical system, to raise the after discharge threshold caused by thalamic and cortical stimulation and to increase the electrical ground activity of the rhinencephalon. A sleep pattern after reserpine was not seen. An action at the thalamic-cortical level is suggested. Rinaldi and Himwich[214] gave doses of up to 0·5 mg. per kg. of reserpine to unanæsthetised, curarised rabbits and could observe no change in the electrical activity of the brain while 1·5 to 2·0 mg. per kg. gave rise to an alertness pattern. Reserpine was thought to stimulate the meso-diencephalic activating system. Chusid and his colleagues[215] have studied the behavioural changes after reserpine administration in epileptic monkeys, and found that these were similar to those observed

in similarly treated human subjects. (A motion picture showing the effects on the behaviour of normal monkeys after reserpine administration has been made by Earl, Dibble and Wolf[216]).

Bein[217] pointed out that reserpine did not exert its influence upon respiration by acting on vagal respiratory reflexes or upon the medullary inspiratory and expiratory centres nor were motor pathways and neuromuscular transmission affected. Schneider and his colleagues[218] showed that 5 mg. per kg. of reserpine facilitated the knee jerk in spinal cats and increased the height of the monosynaptic spike as measured in the ventral root of the spinal cord of the decerebrate cat in which the dorsal spinal root was electrically stimulated. Chen [219] has shown that reserpine facilitates electrically induced hind-leg extensor seizures in mice. It antagonised the actions of phenytoin in a competitive fashion. Reserpine was thought to facilitate seizure spread in the central nervous system. Schneider[220] tested the activity and the responsiveness of diencephalic sympathetic centres in cats, before and after reserpine (0·5 mg. per kg. by intravenous injection or 10 mg. per kg. by mouth). Intact cats showed excitement followed by quiet and some signs of discomfort; there was miosis, salivation, diarrhœa and squinting on exposure to light. After the drug, cats in "sham rage" following hypothalamic transection became quiet and unresponsive and made no efforts to climb or to walk. After intravenous injection of 0·1 mg. per kg. reserpine, a pressor response could still be obtained from direct electrical stimulation of the hypothalamus, but the carotid sinus pressor response was markedly antagonised. Reserpine was considered to have an indirect damping effect on the central nervous system (diencephalic-sympathetic centre), blocking or inhibiting afferent stimuli centrally. In another publication Schneider and Earl[221] have shown that there are quantitative differences between the reactions to reserpine in monkeys and in dogs, cats, rabbits and rats. Jungle-born macaca monkeys became calm and relaxed and even playful under the drug's influence, the EEG being unchanged. The drug in monkeys was thought to act mainly on the brain stem. Recently Brodie and his colleagues[222–225] have given additional confirmation to observations that that lysergic acid diethylamide produces mental disturbances by suppression of some of the central actions of 5-hydroxytryptamine (5-HT). They have noted[222] that 5-HT acts like chlorpromazine and reserpine, in markedly potentiating the hypnotic action of hexobarbitone in mice by increasing the sensitivity of the central nervous system to the hypnotic drug. 5-HT and reserpine were shown to exert a common central potentiating action on hexobarbitone and ethanol in mice[223] which was antagonised by lysergic acid diethylamide. It was suggested that certain actions of reserpine might be due to 5-HT release as indicated by a marked increase in urinary excretion of 5-hydroxyindoleacetic acid in dogs treated with reserpine[223]. Reserpine also released 5-HT from the intestine[224] of rabbits receiving 5 mg. per kg. by intraperitoneal injection. The animals were later killed and the 5-HT content of a portion of the small intestine estimated, using the method of Udenfriend and others[226]. The intestinal 5-HT content fell

to 15 to 20 per cent. of the average normal amount, returning to normal values after 5 days.

It is the general view that the site of action of reserpine is in the brain. Evidence for a peripheral site of action has, however, been given by McQueen, Doyle and Smirk[210]. In the innervated but otherwise isolated hind limb of the rabbit[211] perfused with a blood-dextran medium at a constant rate, there was a rise in limb perfusion pressure (not a fall as might have been expected) after injection of reserpine into the systemic circulation. Direct injection of reserpine into the perfused limb caused a reduction in vasomotor tone. Reserpine had a depressant effect on the actions of vasopressor substances when these were injected into the rat hindquarters preparation and on the response of the isolated rat diaphragm to nervous stimuli. All these reserpine effects were prolonged, suggesting binding of the drug by the musculature. Somewhat similar conclusions have been drawn by Gillis and Lewis[143].

McQueen, Doyle and Smirk[227] have observed cutaneous vasodilatation in the ear vessels of rabbits. This was mainly mediated *via* the nervous system, since, when tested in rabbits which had undergone cervical sympathectomy on the left side together with removal of the superior cervical and stellate ganglia, there was usually dilatation after reserpine in the innervated ear, but no change or constriction in the denervated ear. Gillis and Lewis[143], in studies upon isolated organs, have shown that reserpine (0·1 μg. per ml.) in the isolated perfused kitten's heart caused an increase in outflow followed by a decrease in heart rate and amplitude. The effects were long lasting but usually reversible. One μg. per ml. of reserpine reduced vasoconstriction caused by 100 μg. per ml. barium chloride. Reserpine 1·0 or 0·1 μg. per ml. had little effect upon the response of the heart to adrenaline, noradrenaline or 5-HT. The alkaloid had no direct action upon the isolated guinea-pig ileum, but reduced the responses to acetylcholine, barium chloride, histamine and 5-HT. A delayed maximum inhibitory effect was noted but the responses were not quantitative. Antagonism to the effects of acetylcholine, 5-HT and potassium on the rat's uterus was shown by reserpine, once again with a delayed maximal effect. In the isolated perfused rat hind quarters no direct effect upon outflow was observed with reserpine but the constrictor response to adrenaline was inhibited. Neuromuscular transmission in the isolated sartorius muscle-ischiad nerve preparation of the frog was not modified by the alkaloid, and there was no direct effect upon the isolated frog's rectus abdominis muscle nor upon acetylcholine- or potassium-induced contractions in this tissue.* Reserpine is considered to inhibit some fundamental biochemical contractile process within or upon the surface of the muscle cells.

In the metabolic studies on rats, Sheppard and his colleagues[147] showed a concentration of the drug in fatty tissues which may account for its prolonged action. There is no specific accumulation of reserpine in the

* Since this article went to press a stimulant action on the frog rectus abdominis muscle has been shown [143, 143a].

brain and in view of the somewhat conflicting evidence on the precise site of action this is a significant finding.

A number of other studies have so far been made upon various aspects of the pharmacology of reserpine. For example[228], low doses cause vomiting in pigeons with a graded dose-response relationship within the range of 0·04 to 0·10 mg. per kg. body weight, the use of this response as a possible assay method has been suggested. Barrett and his colleagues[229] have shown that reserpine causes gastric secretion in dogs by stimulation of parasympathetic ganglia. Meier and his colleagues[230] have analysed the actions of a number of hypotensive substances including reserpine. Using anæsthetised rabbits the hypotensive agents were classified on the basis of (*a*) degree of maximal fall of blood pressure, (*b*) duration of the total fall of blood pressure, (*c*) duration of the maximal fall of blood pressure, and (*d*) the action integral which is the product of the intensity and duration of effect. The properties of reserpine were linked with those of hydrallazine since with both drugs the duration of the hypotension was approximately related to the dose.

So far no mention has been made of any influence of reserpine upon the endocrine glands. This subject was investigated in 1954 by Gaunt and his colleagues[231], who found that there was a reduction in fluid intake and urine volume when guinea-pigs were given 3 μg. per 100 g. per day and rats 10 μg. per 100 g. per day. On water-loaded rats, reserpine had an antidiuretic effect but sodium excretion was little changed. No changes in male gonadial function were noticed in the rat unless reserpine was given in doses causing loss of weight and inanition when there was a lowered secretion of androgens. In female rats reserpine (5 or 10 μg. per 100 g. per day) disturbed the normal vaginal œstrous cycle. Reserpine at the lower of these doses reduced the conception rate, number of live births and number of young per litter in rats. Mercier-Parot and Tuchmann-Duplessis[233] have recently shown that 50 μg. per day of reserpine cause disappearance of œstrus in the rat. No evidence was obtained for suppression of ACTH release. Reserpine caused adrenal hypertrophy, and thymic atrophy at a dose of 10 μg. per 100 g. per day. After adrenalectomy in the rat, reserpine reduced survival time suggesting that it increased the need for the cortical hormone and was an adrenal stimulant. Reserpine reduced the blood pressure in DOCA-salt—or cortisone induced hypertension. Sturtevant[232] in 1953 had also produced hypotension in rats with DOCA-salt induced hypertension by the addition of 5 per cent. of *R. serpentina* roots (4 g. of root per day) to their diet.

A thyroxine-induced increase in oxygen consumption was antagonised by reserpine in rats, although it had no influence upon that produced by 2:4-dinitrophenol[234]. Kuschke and Frantz[235] have shown that reserpine causes hyperglycæmia in rabbits without change in sugar tolerance. The hyperglycæmic and sedative effects of reserpine were inhibited by hydergine but bilateral splanchnicotomy did not prevent the hyperglycæmia. Turner and Carl[236] found that reserpine dilates melanophores and lipophores of fish causing display of colour. This effect was inhibited by cocaine and by ephedrine and partly by ergotamine,

but not by phenylephrine, pitocin or thyroid while adrenaline caused blanching.

Before discussing the pharmacology of the other rauwolfia alkaloids it is necessary to refer to the influence of reserpine upon normal human subjects. A very considerable amount of attention has been paid to the influence of reserpine upon hypertensive, psychiatric and other patients and this contrasts with the lack of data available about the normal individual. The study of the effects of reserpine upon the normal ambulant human subject is difficult, since small doses produce little demonstrable effect whilst larger doses of the order of 20 to 50 μg. per kg. may cause unpleasant side effects, including extreme lethargy, drowsiness, shivering, nasal congestion, conjunctival suffusion, weakness and even orthostatic fainting. These effects appear six to twenty-four hours after a single oral or intravenous dose and may persist for several days. There is a slight lowering of the blood pressure, particularly when standing and shortly after meals. Moderate bradycardia is usually present but miosis is not consistently observed. When miosis is present it appears in about twenty-four hours and may persist for twenty-four to forty-eight hours. Plethysmographic studies have shown an increase in hand blood flow which is not necessarily associated with a change in blood pressure[237].

THE PHARMACOLOGY OF OTHER RAUWOLFIA ALKALOIDS

Rescinnamine. The pharmacological properties of this alkaloid are similar to those of reserpine[57,238,239].

Reserpinine. According to Schlittler and his colleagues[82] reserpinine is not sedative or hypotensive. Kroneberg[240] showed that it was neither an adrenergic blocking agent nor spasmolytic, that it potentiated the effects of adrenaline and noradrenaline and caused a fall in blood pressure, but was quantitatively weaker than reserpine.

Serpine. Chatterjee and Bose[65] have described this as a hypotensive drug with a short-lived action. It is an adrenergic blocking agent rather like yohimbine[65]. Dasgupta and Werner[241] have also studied the properties of serpine. In rats, 5 mg. per kg. by intravenous injection cause quietening the LD50 being approximately 10 mg. per kg. Six mg. per kg. produced respiratory distress in monkeys and 7 mg. per kg. inco-ordination and occasional convulsions in rabbits. Serpine induced hypotension in cats and monkeys with suppression of the carotid sinus reflexes. Although an adrenergic blocking agent it did not cause adrenaline reversal. There was some antagonism to the pressor response seen after electrical stimulation of the splanchnic nerves. Serpine caused peripheral vasodilatation in perfused limbs of cats and monkeys, was a weak ganglion blocking agent and stimulated intestinal motility. No specific blockade of the pressor response to electrical stimulation of the medulla and hypothalamus was seen.

Sarpagine-Raupine. Raupine (and raubasin) have been shown by Kroneberg and Achelis[242] to exert adrenergic blocking actions on the blood pressure and nictitating membrane of the cat.

Ajmaline. Ajmaline was shown to have an inhibitory action on frog nerve fibres and ultimately prevented passage of the impulse[243]. Chopra and Chakravarti[245] showed that ajmaline raised the blood pressure of decerebrate cats but caused hypotension in spinal animals. Gupta[246] also observed that ajmaline caused hypertension but that in animals with experimental hypertension it caused a fall in blood pressure, findings confirmed by Chopra and his colleagues[247]. Dasgupta and Werner[248] showed that ajmaline caused a marked fall in blood pressure and inhibition of vasomotor reflexes after intracisternal injection into monkeys. There was reduction in the pressor response to sciatic nerve stimulation in chloralosed and decerebrate cats. Hypertension due to stimulation of the hypothalamus was not suppressed nor was "sham rage" abolished. One to 3 mg. per kg. per day did not cause hypotension in dogs[193].

Serpentine. This alkaloid produces hypotension and inhibits intestinal movements[244,249]. It is more toxic to mice than ajmaline or serpentinine and causes a rise in blood pressure in decerebrate cats, but a fall in spinal cats[245]. It lowers the blood pressure in experimental hypertension[246,247]. It is a more active substance than ajmaline[174]. Its hypotensive activity is probably due to acute vasodilatation[170].

neo*Ajmaline and* iso*ajmaline*[250]. These alkaloids stimulate and then depress the central nervous system in frogs, cats and guinea-pigs. The frog heart *in situ* and isolated rabbit and guinea-pig hearts are depressed. There is peripheral vasodilatation in perfused cat and frog preparations with hypotension in intact, decerebrate and spinal cats. In experimental hypertension *neo*ajmaline and *iso*ajmaline reduced the blood pressure. Respiratory depression is produced by large doses. Death is caused by respiratory failure.

Serpentinine. Serpentinine diminishes the renal vasoconstrictor activity of adrenaline but does not alter adrenaline hypertension[251]. It has hypotensive activity[252].

Ajmalicine. Ajmalicine was shown not to have hypotensive activity in dogs by Cronheim, Stipp and Brown[193].

Ajmalinine. Ajmalinine causes hypotension with renal vasodilatation. It is a sympatholytic[253,254].

*R.S.*51[116]. This lowers blood pressure in normo-tensive and hypertensive animals. It is a peripheral vasodilator, an adrenergic blocking agent and a histamine liberator. Hypotension is not due to an action on brain centres, sympathetic ganglia, the myocardium or the splanchnic blood vessels.

Rauwolfinine[98]. This alkaloid has hypertensive properties.

Rauwolfine. This alkaloid may be identical with ajmaline but will be given separate, brief consideration. Rauwolfine was found to increase myocardial resistance to fibrillation in dogs and cats. De Boer[256,257] showed that it decreased the rate of the frog, cat and rabbit heart, and caused artificial changes in rhythm with depression of intraventricular conduction. It did not prevent auricular or ventricular fibrillation in cats or rabbits. Studies of the effects of rauwolfine on frog and mammalian hearts have also been made by Hartog[258].

THE ALKALOIDS OF OTHER *Rauwolfia* SPECIES

R. canescens[259]

Rauwolscine. This is a cardiovascular depressant with hypnotic activity and a relatively high toxicity[260]. It causes hypotension, blocks the effects of adrenaline on the cat blood pressure and antagonises the action of adrenaline on the mammalian and amphibian heart[261,262]. Mukherjee and Sen[263–265], have observed a reversible depressant effect upon the toad heart. There was an increase in coronary flow in the perfused guinea-pig and rabbit hearts when the perfusing fluid contained 10 μg. per ml. of rauwolscine; a solution of 1 mg. per ml. caused irreversible standstill. Rauwolscine caused hypotension in the normal cat with inhibition of cardiac and plain muscle, blocking of the vagus and antagonism to the pressor effects of adrenaline. Werner and his colleagues[266,267] gave rauwolscine by intra-cisternal injection to monkeys and showed that it caused hypotension with inhibition of the carotid sinus pressor reflexes. It is a vasodilator and has adrenergic blocking activity at high dose levels. Rauwolscine, when injected into the trunk of an animal, causes vasoconstriction of the innervated but otherwise isolated hind limb. In cats and monkeys there is a marked fall in peripheral resistance without alteration of cardiac output.

Canescine, Recanescine, Deserpidine[268–271]. These appear to be identical substances with properties very similar to those of reserpine and rescinnamine. The CH_3O group at the 11-position in reserpine does not seem to be essential for its pharmacological actions.

R. hirsuta (*R. heterophylla*)[131,272]

The *R. hirsuta* alkaloids are pharmacologically active and are described as "rautensin" by Mezey and Uribe[131,272]. This substance is probably a mixture of alkaloids. It causes in dogs and cats apathy, adynamia, hypotension, bradycardia, ECG changes and shortness of breath. Death is caused by respiratory failure.

R. sellowii[134]

Aqueous extracts are toxic to mice. The total alkaloids cause hypotension.

R. vomitoria[273]

Crude aqueous extracts prepared from the roots have been used. In the bilaterally vagotomised dog, these caused inhibition and reversal of the pressor response to adrenaline with less inhibition of the tone and movement of intestinal smooth muscle. The extracts themselves caused a rise in blood pressure and an increase in the tone and amplitude of movements of the small intestine.

SYNTHETIC AND NATURALLY OCCURRING COMPOUNDS RELATED TO THE RAUWOLFIA ALKALOIDS

Rauwolfine[274]. This is obtained from *Tabernæmontana ventricosa* Hocht (Apocynaceæ) which grows in South Africa. It has sympatholytic properties.

Esters of Yohimbine and Corynanthine. Le Hir[275] has studied the 3:4:5-trimethoxybenzoic acid esters of yohimbine and corynanthine and compared their properties with those of reserpine. In mice these esters were found to be less toxic than reserpine. They did not augment the hypnotic effects of amylobarbitone. The yohimbine derivative caused hypotension and adrenaline reversal, with a diminished response to carotid sinus occulsion. The corynanthine ester had somewhat similar properties. Huebner and his colleagues[276] have prepared the following yohimbé alkaloid derivatives and shown that they lack the characteristic sedative actions of reserpine:—

o-benzoyl, -anisoyl, -veratroyl and 3:4:5-trimethoxybenzoylyohimbine,
o-3:4:5-trimethoxybenzoylcorynanthine,
o-3:4:5-trimethoxybenzoyl-α-yohimbine,
o-acetylyohimbine, yohimbic acid amide and hydrazide, *N*-methyl yohimbine.

Discussion

Only the pharmacological properties and mode of action of reserpine will be considered. Reserpine is an intriguing drug. Its interest lies very much in its apparent lack of specificity and selectivity of action. If appropriate steps are taken, then a response to reserpine administration can be obtained from almost all systems, tissues and organs. This response is rarely a dramatic one, compared with the effects of say adrenaline or histamine, and it usually takes a little time to appear and then it is relatively persistent. In some cases, changes are not evident for up to one year as has been described in the onset of Parkinsonism. Opinion on the site of action of reserpine favours the brain and central nervous system as being the primary areas affected, and the rest of the organism being secondarily influenced. This may well be so, but there is good evidence for the direct action of the drug on other tissues. It is possible that the best explanation of the mode-of-action of reserpine is that it exerts a depressant effect upon some ubiquitous biochemical system in the cells, which is concerned with respiration or with some other aspect of the cell's economy. This might explain the delayed action since the effects of depression of metabolite synthesis are not always seen at once. In addition it is conceivable that this same, admittedly hypothetical, effect would, when exerted in nervous tissue, produce more profound and readily demonstrable effects than when exerted elsewhere.

It is interesting to speculate upon the part played by 5-HT in the central nervous actions of reserpine. The possibility that this substance is involved in mental activity has been discussed by Gaddum[277] and by Woolley and Shaw[278]. 5-HT has been found in the brain, and its distribution is both interesting and suggestive[279,280]. It has been postulated that the central effects of reserpine are mediated by liberation of 5-HT[222–226] since both depress mice and potentiate the actions of hexobarbitone in these animals. These potentiatory effects are antagonised by lysergic acid diethylamide. The antagonism between reserpine and lysergic acid diethylamide may point to an action of the former being mediated by

5-HT. Recently, however, Cerletti and Rothlin[281] have investigated the actions of 2-brom-(+)-lysergic acid (BOL 148) and have shown that this powerful peripheral antagonist to 5-HT does not produce the mental aberrations characteristic of lysergic acid diethylamide. This finding at once throws open the question about the part played by 5-HT in the brain. It would, however, be interesting to know how the distribution of 5-HT (and noradrenaline) in the brain compares with that of reserpine, in a well sedated animal.

Bein[217] in a recent paper has discussed the mode of action of reserpine with particular reference to its influence upon central reflex mechanisms. He has considered the behaviour of central autonomic efferent impulses after afferent nerve stimulation. Inhibition of the carotid sinus pressor reflex in the cat by reserpine has been shown to depend upon a connection being maintained between the medulla and the mid-brain. Bein has also discussed the influence of reserpine upon respiration. Spontaneous respiration is influenced by reserpine in a characteristic manner which is in some ways analogous to its action upon the carotid sinus pressor reflex. It is felt that reserpine may activate inhibiting centres in the brain which probably lie rostral to the caudal colliculi. It acts upon central regulatory mechanisms which integrate both somatic and autonomic functions[217].

Tripod and Meier[198] account for the direct and antagonistic actions of reserpine upon the isolated blood vessels of the rabbit hind quarters and upon the coronary vessels of the isolated mammalian heart by postulating the possible existence of "master receptors", which are different from those which subserve adrenergic, cholinergic and histaminergic functions. These determine the order of priority of the primary effects of drug upon cell, and also its antagonisms.

In conclusion it appears that we are still some way from knowing exactly at what point in the metabolism of the cell reserpine acts, nor do we know precisely its mode of action. It seems not improbable that the biochemist and not the pharmacologist will provide the final answer. It is possible that the action is upon the contractile substance of muscle cells, and it may be exerted upon some step in the synthetic, degradative or oxidative processes which go on in all cells. This may not, of course, explain the mental effects of the drug or its influence upon nervous tissues.

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