

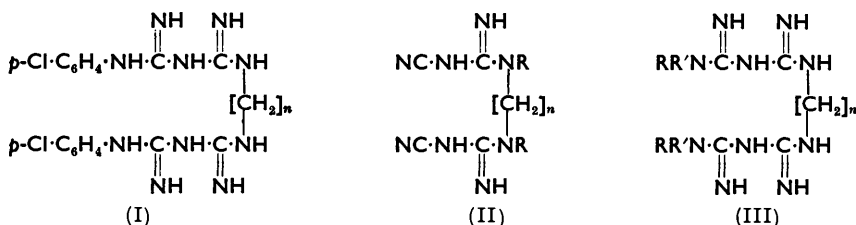
850. *Bisdiguanides having Antibacterial Activity.*

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A series of bisdiguanides has been prepared either by condensing one mol. of a biscyanoguanidine with two mols. of an amine hydrochloride, or by the converse interaction of two mols. of an *N*-arylcyanoguanidine with one of a diamine salt. One of the products, 1:6-di-(*N*<sup>5</sup>-*p*-chlorophenyl-*N*<sup>1</sup>-diguanido)hexane (chlorohexidine B.P.C.), has found practical use as an antibacterial agent.

THE occurrence of antimalarial activity in certain substituted diguanides<sup>1</sup> stimulated the search for other therapeutically useful members of this series, and in due course led to the discovery of high antibacterial activity, more especially amongst a series of bisdiguanides. One such compound, 1:6-di-(*N*<sup>5</sup>-*p*-chlorophenyl-*N*<sup>1</sup>-diguanido)hexane (I; *n* = 6) has recently been introduced into medical and veterinary practice under the common name chlorohexidine B.P.C. ("Hibitane" \*).<sup>2</sup> The present communication is concerned with the chemistry of this substance, and also with that of related compounds, many of which were prepared during the investigation of its mode of action.

The original observation of marked bacteriostatic action was made with the mixture of polymeric diguanides that resulted from the fusion of 1:6-di-(*N*<sup>3</sup>-cyano-*N*<sup>1</sup>-guanidino)hexane (II; R = H, *n* = 6) with hexamethylenediamine dihydrochloride. Attempts to



determine actual chain lengths, or to separate the mixture into homogeneous fractions, were unsuccessful. Since, however, it was known that molecules carrying only one diguanide residue were but weakly antibacterial, it was clearly desirable to determine the degree of molecular complexity necessary for high antimicrobial potency. For this purpose the step-wise synthesis of polydiguanides was undertaken. In the event, full biological activity was reached immediately two diguanide systems were incorporated into each drug molecule, and effort was then concentrated on ascertaining the optimum distance which should exist between these two residues and the most effective types of end groupings—whether, for example, antibacterial activity was highest in bisdiguanides in which the terminal groups were aryl, alkyl, or heterocyclic. In addition it was desired to examine the effect of replacing the diguanide residues by other less complex basic groups such as that of guanidine. The relevant biological findings have already been reported elsewhere;<sup>3</sup> briefly it was found that highest antibacterial activity occurred in the series (I), and in particular when *n* = 5, 6, or 7. Since hexamethylenediamine was most readily available to us, the majority of the subsequent preparations were based on this substance. The introduction of hydroxyl or carbonyl substituents into the terminal aryl groups almost eliminated antibacterial action, as did the replacement of diguanide by guanidine, while the analogous wholly alkyl-bisdiguanides (III; R = alkyl, R' = H or alkyl, *n* = 6) were only fractionally as effective (one-third to one-tenth) as the compound (I).

The standard methods of preparing the bisdiguanides were employed, similar to those developed for the simple antimalarial monodiguanides.<sup>1,4</sup> The first required the reaction

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<sup>1</sup> Curd and Rose, *J.*, 1946, 729.

<sup>2</sup> B.P. 705,838.

<sup>3</sup> Davies, Francis, Martin, Rose, and Swain, *Brit. J. Pharmacol.*, 1954, 9, 192.

<sup>4</sup> Curd, Hendry, Kenny, Murray, and Rose, *J.*, 1948, 1630.

TABLE 1. *Biscyanoguanidines* (II) (R = H except as stated).

Code no.	-[CH <sub>2</sub> ] <sub>n</sub> -	Butanol (c.c.) / 0.01 mol.	Reaction time (hr.)	Solvent for crystn.	M. p. †	Yield (%)	Formula	Found (%)			Required (%)		
								C	H	N	C	H	N
10,184	[CH <sub>2</sub> ] <sub>2</sub>	15	12	H <sub>2</sub> O	248-250° †	57	C <sub>6</sub> H <sub>10</sub> N <sub>8</sub>	36.35	5.3	56.8	37.1	5.15	57.7
10,188	[CH <sub>2</sub> ] <sub>3</sub>	15	16	"	206 †	50-60	C <sub>7</sub> H <sub>12</sub> N <sub>8</sub>	39.95	5.9	53.5	40.4	5.8	53.5
11,108	[CH <sub>2</sub> ] <sub>4</sub>	14	18	"	197-199	74	C <sub>8</sub> H <sub>14</sub> N <sub>8</sub>	43.0	6.9	48.1	43.2	6.3	50.4
10,689	[CH <sub>2</sub> ] <sub>4</sub>	14	20	"	169-171	58	C <sub>8</sub> H <sub>14</sub> N <sub>8</sub>	45.2	6.3	46.6	45.8	6.8	47.45
10,691	[CH <sub>2</sub> ] <sub>5</sub>	14	8	"	202-203 (s 199)	70-80	C <sub>10</sub> H <sub>18</sub> N <sub>8</sub>	47.45	7.45	44.4	48.0	7.2	44.8
10,940	[CH <sub>2</sub> ] <sub>5</sub>	12.5	16	Aq. EtOH	144-146 (s 138-140)	30-40	C <sub>11</sub> H <sub>20</sub> N <sub>8</sub> ·H <sub>2</sub> O	48.25	7.55	41.25	48.3	7.7	41.0
11,380	[CH <sub>2</sub> ] <sub>5</sub>	15	"	* Aq. EtO·C <sub>2</sub> H <sub>4</sub> ·OH	184-186 (s 182)	v. low	C <sub>11</sub> H <sub>20</sub> N <sub>8</sub>	54.15	8.0	36.6	54.9	8.5	36.6
11,386	[CH <sub>2</sub> ] <sub>5</sub>	15	"	* Aq. EtOH	186-187	83	C <sub>11</sub> H <sub>20</sub> N <sub>8</sub>	59.3	9.45	30.85	59.7	9.4	30.9
11,382	[CH <sub>2</sub> ] <sub>5</sub>	10	"	* H <sub>2</sub> O	138-140 (s 136)	56	C <sub>10</sub> H <sub>18</sub> N <sub>8</sub>	44.45	6.65	42.3	45.1	6.8	42.1
11,384	[CH <sub>2</sub> ] <sub>5</sub>	15	15	* Aq. EtO·C <sub>2</sub> H <sub>4</sub> ·OH	204-206	56	C <sub>11</sub> H <sub>20</sub> N <sub>8</sub>	53.25	6.35	30.1	53.6	6.1	31.3
10,388	[CH <sub>2</sub> ] <sub>5</sub>	10	16	Gum used without further purification after removal of salt and solvent.									

\* Prisms. † s = sinters. ‡ With decomp. § R = Bu<sup>l</sup>.

TABLE 2. *Bis-p-chlorophenyldiguanidines* (I).

Code no.	-[CH <sub>2</sub> ] <sub>n</sub> -	2-Ethoxyethanol per 0.01 mol.	Reaction time (hr.)	Solvent for crystn.	M. P.	Yield (%)	Formula	Found (%)			Required (%)			
								C	H	N	C	H	N	
20,184	[CH <sub>2</sub> ] <sub>2</sub>	14	6	H <sub>2</sub> O	245-246° *	26	C <sub>18</sub> H <sub>22</sub> N <sub>10</sub> Cl <sub>2</sub> ·2HCl·H <sub>2</sub> O	40.2	5.2	26.3	25.9	40.0	4.8	25.9
12,483	[CH <sub>2</sub> ] <sub>3</sub>	20	4	"	224-226 *	50	C <sub>18</sub> H <sub>22</sub> N <sub>10</sub> Cl <sub>2</sub> ·2HCl·H <sub>2</sub> O	41.4	5.3	24.5	25.2	41.15	5.05	25.3
12,484	[CH <sub>2</sub> ] <sub>4</sub>	20	3	"	253-254 *	59	C <sub>20</sub> H <sub>26</sub> N <sub>10</sub> Cl <sub>2</sub> ·2HCl	43.7	5.2	24.6	25.2	43.6	5.1	25.5
12,485	[CH <sub>2</sub> ] <sub>5</sub>	20	6	"	250	58	C <sub>21</sub> H <sub>28</sub> N <sub>10</sub> Cl <sub>2</sub> ·2HCl	44.4	5.2	24.2	25.5	44.7	5.3	25.8
10,040	[CH <sub>2</sub> ] <sub>6</sub>	See text		"										
12,486	[CH <sub>2</sub> ] <sub>6</sub>	25	1½	"	253-254	57	C <sub>23</sub> H <sub>32</sub> N <sub>10</sub> Cl <sub>2</sub> ·2HCl	46.0	5.3	23.7	25.0	46.6	5.7	23.6
11,383	[CH <sub>2</sub> ] <sub>10</sub>	20	6	Aq. EtOH	246-248	60	C <sub>26</sub> H <sub>38</sub> N <sub>10</sub> Cl <sub>2</sub> ·2HCl	49.25	6.3	21.4	—	49.2	6.3	22.1
14,345	[CH <sub>2</sub> ] <sub>10</sub>	18	2	H <sub>2</sub> O	206-238	73	C <sub>23</sub> H <sub>30</sub> ON <sub>10</sub> Cl <sub>2</sub> ·2HCl	44.4	5.5	23.3	24.0	44.4	5.4	23.6
14,411	[CH <sub>2</sub> ] <sub>13</sub>	20	2½	Aq. COMe <sub>2</sub>	204-205	50	C <sub>23</sub> H <sub>30</sub> N <sub>10</sub> Cl <sub>2</sub> ·2HCl	46.7	5.4	22.6	22.2	45.1	5.6	22.0
11,382	[CH <sub>2</sub> ] <sub>13</sub>	20	9	Aq. EtOH	248-249	35	C <sub>28</sub> H <sub>34</sub> O <sub>2</sub> N <sub>10</sub> Cl <sub>2</sub> ·2HCl	49.05	5.2	20.4	—	49.0	5.3	20.4
11,384	[CH <sub>2</sub> ] <sub>13</sub>	—	—	"	253-254	70	C <sub>28</sub> H <sub>32</sub> N <sub>10</sub> Cl <sub>2</sub> ·2HCl	46.15	4.35	24.7	—	46.3	4.2	24.6
11,385	(p)C <sub>6</sub> H <sub>4</sub> ·CH <sub>2</sub> ·C <sub>6</sub> H <sub>4</sub> (p)†	—	—	"	—	50	C <sub>29</sub> H <sub>32</sub> N <sub>10</sub> Cl <sub>2</sub> ·2HCl	52.85	4.6	20.4	—	52.7	4.5	21.2

\* With decomp. † Method (ii), see text.

TABLE 3. 1 : 6-*Bisdiguanidohexanes* (III); R' = H, n = 6).

Code no.	R	2-Ethoxyethanol (c.c.) / 0.01 mol.	Reaction time (hr.)	Solvent	M. P.	Yield (%)	Formula	Found (%)			Required (%)			
								C	H	N	C	H	N	
10,387	Ph <sup>b</sup>	16	3	H <sub>2</sub> O	242-244°	74	C <sub>22</sub> H <sub>32</sub> N <sub>10</sub> ·2HCl	51.55	6.7	27.4	12.9	51.9	6.7	27.5
10,388	p-C <sub>6</sub> H <sub>4</sub> Me	25	2	Aq. EtO·C <sub>2</sub> H <sub>4</sub> ·OH	263-264	90	C <sub>24</sub> H <sub>36</sub> N <sub>10</sub> ·2HCl	53.45	6.5	25.65	—	53.6	7.1	26.1
11,108	p-C <sub>6</sub> H <sub>4</sub> ·OH <sup>c</sup>	"	2½	EtOH-PrOH	200-202	40	C <sub>22</sub> H <sub>32</sub> O <sub>2</sub> N <sub>10</sub> ·2HCl	48.4	6.15	25.3	—	48.8	6.3	25.9
10,689	p-C <sub>6</sub> H <sub>4</sub> ·OMe <sup>b</sup>	"	2	MeOH-EtOH	238-240	78	C <sub>24</sub> H <sub>34</sub> O <sub>2</sub> N <sub>10</sub> ·2HCl	49.85	6.5	25.5	—	50.6	6.7	24.6
10,691	p-C <sub>6</sub> H <sub>4</sub> ·CO <sub>2</sub> H	25	6	H <sub>2</sub> O	218-224 *	42	C <sub>24</sub> H <sub>36</sub> O <sub>2</sub> N <sub>10</sub> ·2HCl	47.65	6.1	22.7	—	48.2	5.7	23.4
10,940	p-C <sub>6</sub> H <sub>4</sub> Cl <sup>c</sup>	25	3	"	260-262	90	C <sub>22</sub> H <sub>30</sub> N <sub>10</sub> Cl <sub>2</sub> ·2HCl	45.5	5.8	24.25	23.8	45.7	5.5	24.2
11,380	m-C <sub>6</sub> H <sub>4</sub> Cl	25	3	Aq. EtOH	233-234	10	C <sub>22</sub> H <sub>30</sub> N <sub>10</sub> Cl <sub>2</sub> ·2HCl	46.3	5.95	23.95	—	46.7	4.5	24.2
11,386	3 : 4-C <sub>6</sub> H <sub>3</sub> Cl <sub>2</sub>	13	9	EtO·C <sub>2</sub> H <sub>4</sub> ·OH	259-260	68	C <sub>22</sub> H <sub>28</sub> N <sub>10</sub> Cl <sub>2</sub> ·2HCl	41.2	5.0	21.5	—	40.8	4.6	21.6
11,381	2 : 5-C <sub>6</sub> H <sub>3</sub> Cl <sub>2</sub>	25	8	Aq. EtOH	249-250	34	C <sub>22</sub> H <sub>28</sub> N <sub>10</sub> Cl <sub>2</sub> ·2HCl	40.6	5.2	20.9	—	40.8	4.6	21.6
11,110	α-C <sub>10</sub> H <sub>7</sub>	"	8	Aq. AcOH	257-258	48	C <sub>30</sub> H <sub>38</sub> N <sub>10</sub> ·2HCl	58.9	6.1	22.7	—	59.1	5.9	23.0
10,388	β-C <sub>10</sub> H <sub>7</sub> <sup>b</sup>	"	3	H <sub>2</sub> O	252-254	74	C <sub>30</sub> H <sub>38</sub> N <sub>10</sub> ·2HCl	59.5	6.2	23.1	10.9	59.1	5.9	23.0

<sup>a</sup> Prisms. <sup>b</sup> Needles. <sup>c</sup> Plates. \* With decomp.

in nitrobenzene of an *N*-phenylcyanoguanidine with each of the amino-groups of a diamine, and the second, more generally used, was the complementary process in which the biscyanoguanidine (II) interacted with two molecular proportions of the amine NHRR' (see III), preferably in boiling 2-ethoxyethanol. The biscyanoguanidines were themselves readily accessible from the reaction of the appropriate diamine dihydrochloride with two molecular proportions of sodium dicyanimide in boiling butanol. The yield of the crystallised products was 50—80%, that of the hexamethylene derivative being in the upper range. The intermediate preparations also included examples in which the nitrogen atoms adjacent to the polymethylene chain carried an alkyl group (II; R = alkyl), and others in which ether-oxygen atoms, and in one case additionally a *p*-substituted phenyl residue, were interposed in the polymethylene chain (see Table 1).

A slightly modified procedure was employed for the production of the bisdiguanydes (III; R and/or R' = alkyl) in that a solvent was not required, and the usual practice was to fuse an intimate mixture of the biscyanoguanidines with the alkylamine hydrochlorides at bath temperatures in the range 150—160°. Two bisdiguanydes were also made in which the terminal groups were derived from 8-amino-6-methoxyquinoline and 2:5-diamino-4:6-dimethylpyrimidine severally: in these instances, 2-ethoxyethanol was used as solvent. Finally, the bisguanidine corresponding to the bisdiguanyde (I; *n* = 6) was prepared from the related thiourea, itself formed by the interaction of hexamethylene diisothiocyanate with *p*-chloroaniline in boiling ethanol. Amination was effected by alcoholic ammonia in the presence of mercuric oxide.

#### EXPERIMENTAL

*General Method of Preparation of Biscyanoguanidines.*—The diamine dihydrochloride (1.0 mol.) and sodium dicyanimide (2.0 mols.) were powdered together, mixed with butanol, and stirred and heated under reflux (bath-temp. 130—140°) for 8—16 hr. After cooling, either the insoluble solid was filtered off and washed with cold water to remove sodium chloride, or, where the product remained in solution, the sodium chloride was first filtered off and the filtrate was then evaporated to dryness under reduced pressure. Details of the experiments are given in Table 1.

1:6-Di-(*N*<sup>6</sup>-*p*-chlorophenyl-*N*<sup>1</sup>-diguanydo)hexane Dihydrochloride (10,040).—(i) 1:6-Di-(*N*<sup>3</sup>-cyano-*N*<sup>1</sup>-guanidino)hexane (30 g., 0.12 mol.), *p*-chloroaniline hydrochloride (39.6 g., 0.12 mol.), and 2-ethoxyethanol (300 c.c.) were stirred together under reflux (bath-temp. 130—140°) for 3 hr. After cooling, the microcrystalline *dihydrochloride* [62 g., 90%; m. p. 260—262° (decomp.)] was filtered off and washed with ethanol. Recrystallisation from water, in which it is sparingly soluble, gave plates, m. p. unchanged (Found: C, 45.5; H, 5.8; N, 24.25; Cl, 23.8. C<sub>22</sub>H<sub>30</sub>N<sub>10</sub>Cl<sub>2</sub>·2HCl requires C, 45.7; H, 5.5; N, 24.2; Cl, 24.9%). The *base* could be obtained sensibly free from the hydrochloride only by adding a solution of the latter in hot water to an excess of aqueous sodium hydroxide, and not by the reverse procedure. It formed colourless needles (from methanol), m. p. 133.5—134° (corr.) (Found: C, 52.1; H, 6.0; N, 27.7. C<sub>22</sub>H<sub>30</sub>N<sub>10</sub>Cl<sub>2</sub> requires C, 52.3; H, 5.9; N, 27.7%). The *diacetate* crystallised on cooling from a solution of the *base* in hot dilute acetic acid and after recrystallisation from water formed prisms, m. p. 154—155° (corr.) (Found: C, 48.6; H, 6.4; N, 21.7; loss at 120° *in vacuo*, 3. C<sub>22</sub>H<sub>30</sub>N<sub>10</sub>Cl<sub>2</sub>·2C<sub>2</sub>H<sub>4</sub>O<sub>2</sub>·H<sub>2</sub>O requires C, 48.5; H, 6.2; N, 21.75; H<sub>2</sub>O, 2.8%).

(ii) *N*<sup>3</sup>-*p*-Chlorophenyl-*N*<sup>1</sup>-cyanoguanidine (19.5 g., 0.1 mol.), hexamethylenediamine dihydrochloride (10.4 g., 0.11 mol.), and nitrobenzene (50 c.c.) were stirred together at 140—145° (bath-temp.) for 16 hr. The suspension was filtered hot and the well-pressed filter cake was suspended in ethanol and stirred for 1½ hr. Refiltration gave the crude dihydrochloride (15.3 g., 53%; m. p. 257—258°). Substitution of methylcyclohexanol for nitrobenzene gave only a 20% yield.

All the bis-*p*-chlorophenyldiguanydes listed in Tables 2 and 3, with two exceptions, were prepared by method (i). The two exceptions, 11,384 and 11,385 (Table 2), were prepared by method (ii).

*Miscellaneous Bisdiguanydes.*—1:6-Di-(*N*<sup>6</sup>-*n*-butyl-*N*<sup>1</sup>-diguanydo)hexane dihydrochloride (9458). 1:6-Di-(*N*<sup>3</sup>-cyano-*N*<sup>1</sup>-guanidino)hexane (1.25 g., 0.055 mol.) and *n*-butylamine hydrochloride (1.1 g., 0.01 mol.) were powdered together and heated at 150—155° for 2 hr. The clear melt solidified after 1½ hr., and the *product* was obtained as a microcrystalline solid

(1.4 g., 59.6%; m. p. 223—224° with sintering at 190—192°) by addition of ethyl acetate to a solution in ethyl alcohol (Found: C, 45.5; H, 8.55; N, 29.55; Cl, 15.15.  $C_{18}H_{40}N_{10} \cdot 2HCl$  requires C, 46.1; H, 8.95; N, 29.85; Cl, 15.1%). The salt was freely soluble in water.

1:6-Di-( $N^5$ -isopropyl- $N^1$ -diguanido)hexane dihydrochloride (9357). 1:6-Di-( $N^5$ -cyano- $N^3$ -guanidino)hexane (1.25 g., 0.005 mol.) and isopropylamine hydrochloride (0.9 g., 0.01 mol.) were intimately mixed and heated together at 160° for  $\frac{1}{2}$  hr. Similarly crystallised from alcohol-ethyl acetate, the microcrystalline dihydrochloride (1.1 g., 50%) had m. p. 256—257° (decomp.) (Found: C, 43.3; H, 8.3; N, 32.3.  $C_{16}H_{36}N_{10} \cdot 2HCl$  requires C, 43.5; H, 8.6; N, 31.75%) and was freely soluble in water.

1:6-Di-( $N^5$ -cyclohexyl- $N^1$ -diguanido)hexane dihydrochloride (9382). 1:6-Di-( $N^3$ -cyano- $N^1$ -guanidino)hexane (1.25 g., 0.005 mol.) and cyclohexylamine hydrochloride (1.35 g., 0.01 mol.) were intimately mixed and heated at 160° for 1 hr. Similarly recrystallised from alcohol-ethyl acetate, the dihydrochloride had m. p. 234—236° after sintering at 225—230° (Found: N, 26.25.  $C_{22}H_{44}N_{10} \cdot 2HCl$  requires N, 26.9%).

1:6-Di-( $N^5N^5$ -pentamethylene- $N^1$ -diguanido)hexane dihydrochloride (9383). 1:6-Di-( $N^3$ -cyano- $N^1$ -guanidino)hexane (1.25 g., 0.005 mol.) and piperidine hydrochloride (1.2 g., 0.01 mol.) were intimately mixed and heated at 150° for  $1\frac{1}{2}$  hr. The originally clear melt solidified after 1 hr. Similarly recrystallised from ethanol-ethyl acetate, the dihydrochloride formed prisms, m. p. 231° (1.1 g., 45%) (Found: N, 28.6.  $C_{20}H_{40}N_{10} \cdot 2HCl$  requires N, 28.4%).

1:6-Di-[ $N^6$ -(2-amino-4:6-dimethylpyrimidin-5-yl)- $N^1$ -diguanido]hexane disulphate (10,160). 2-Amino-4:6-dimethylpyrimidine (5.6 g., 0.04 mol.) and 2-ethoxyethanol (50 c.c.) and 1:6-di-( $N^3$ -cyano- $N^1$ -guanidino)hexane (5.0 g., 0.02 mol.) added to the stirred solution, followed by ethanolic hydrochloric acid (4.8N; 8.5 c.c., 2 equivs.). The mixture was stirred under reflux for 68 hr., the 2-ethoxyethanol removed under reduced pressure, and the residue dissolved in water (50 c.c.). The brown gum precipitated by addition of 10N-sodium hydroxide was washed by decantation with water and redissolved in dilute hydrochloric acid; the solution was made alkaline with ammonia and filtered (charcoal), and the base reprecipitated with sodium hydroxide solution. After being further washed with water by decantation, the still gummy base was converted into the disulphate (7 g.) by adding successively sulphuric acid ( $d$  1.84; 3 c.c.) and diethylamine to alkalinity, to its solution in 1:1 aqueous ethanol (100 c.c.), filtering (charcoal), and finally adding excess of dry ethanol. Recrystallisation from water afforded prisms (1.8 g.), m. p. 220—225° (decomp.) (Found: C, 35.05; H, 6.4; N, 29.15; S, 8.2.  $C_{22}H_{38}N_{16} \cdot 2H_2SO_4 \cdot 2H_2O$  requires C, 34.8; H, 6.1; N, 29.5; S, 8.4%).

1:6-Di-[ $N^5$ -(6-methoxyquinolin-8-yl)- $N^1$ -diguanido]hexane dihydrochloride (10,690). 8-Amino-6-methoxyquinoline (7.0 g., 0.04 mol.), 1:6-di-( $N^3$ -cyano- $N^1$ -guanidino)hexane (5.0 g., 0.02 mol.), 2-ethoxyethanol (50 c.c.), and ethanolic hydrochloric acid (6.2N; 6.5 c.c., 2 equivs.) were stirred together under reflux for 3 hr. The orange-coloured suspension slowly dissolved to a clear solution, which finally deposited crystals of the dihydrochloride (5.1 g., 45%; m. p. 246—248°). This recrystallised from water in colourless felted needles, m. p. 246—248° (Found: C, 53.2; H, 5.95; N, 24.8.  $C_{30}H_{38}O_2N_{12} \cdot 2HCl$  requires C, 53.65; H, 5.95; N, 25.0%).

1:6-Di-( $N^3$ -*p*-chlorophenyl- $N^1$ -thioureido)hexane (11,368). A mixture of hexamethylene diisothiocyanate (10.0 g., 0.05 mol.), *p*-chloroaniline (13.0 g., 0.05 mol.), and ethanol (150 c.c.) was refluxed for 16 hr. and water was then added to crystallisation point. Cooling gave the crude thiourea (16 g.) which recrystallised from ethanol in colourless needles (13.8 g., 60%; m. p. 171—172°) (Found: C, 53.1; H, 5.25; N, 12.55.  $C_{20}H_{24}N_4S_2Cl_2$  requires C, 52.75; H, 5.3; N, 12.3%).

1:6-Di-( $N^3$ -*p*-chlorophenyl- $N^3$ -guanido)hexane (11,717). Ammonia in ethanol (5.6N; 150 c.c.), followed by mercuric oxide (4.5 g.), was added to a cooled solution of the above thiourea (9.1 g.) in 2-ethoxyethanol (80 c.c.). The suspension was kept at 30—35° for 40 hr., then warmed to 60° and filtered. The bulk of the solvent was distilled under reduced pressure and the guanidine (3.5 g.; m. p. 146—147°) was precipitated by slow addition of water. It crystallised from ethanol-water in colourless needles (2.6 g.; m. p. 160—161°) (Found: C, 57.0; H, 6.25; N, 19.8.  $C_{20}H_{26}N_6Cl_2$  requires C, 57.0; H, 6.2; N, 19.95%).