literature has revealed a number of Mn^{3+} oxides which have similar distorted structures, for example, $Mn^{2+}Mn_2^{3+}O_4$ (hausmannite)[20], $ZnMn_2O_4$ (hetærolite)[20], γ-Mn_2O_3 [21] and MnO(OH) (manganite)[22]. These results confirm our prediction[7] that a large Jahn–Teller distortion should occur for the d^4 configuration. Distortions from cubic symmetry are either absent or extremely small in octahedrally co-ordinated oxides of metal ions which have 3, 5, 6, 7 or 8 *d*-electrons. This also is in agreement with the theory. Two further examples of complexes containing 4 and 9 *d*-electrons respectively, which become distorted in the way which our theory predicts, have just been reported. Manganic fluoride[23], MnF_3, is a distorted form of the VF_3-type transition metal trifluoride structure[24], while copper fluoride, CuF_2, has a distorted rutile-type structure[25] in contrast to the undistorted rutile structures[26] of MnF_2, FeF_2, CoF_2 and NiF_2 [26].

[1] Felsenfeld, G., *Proc. Roy. Soc.*, A, **236**, 506 (1956).
[2] Pauling, L., "The Nature of the Chemical Bond", 104 (2nd edit.) (Cornell Univ. Press, 1940).
[3] Wells, A. F., "Structural Inorganic Chemistry", 622 (2nd edit.) (Oxford Univ. Press, 1950).
[4] Bleaney, B., and Stevens, K. W. H., "Rep. Prog. Phys.", **16**, 108 (1953). Bowers, K. D., and Owen, J., "Rep. Prog. Phys.", **18**, 304 (1955).
[5] Bjerrum, J., Ballhausen, C. J., and Jorgensen, C. I., *Acta Chem. Scand.*, **8**, 1275 (1954).
[6] Orgel, L. E., *J. Chem. Phys.*, **23**, 1004 (1955).
[7] Orgel, L. E., *J. Chem. Soc.*, 4756 (1952); Proc. 10th Solvay Conference in Chemistry, Brussels (1956).
[8] Jahn, H. A., and Teller, E., *Proc. Roy. Soc.*, A, **161**, 220 (1937).
[9] Bleaney, B., and Bowers, K. D., *Proc. Phys. Soc.*, A, **65**, 667 (1952).
[10] Van Vleck, J. H., *J. Chem. Phys.*, **7**, 472 (1939).
[11] Vainstein, B. I., *Doklady Akad. Nauk, S.S.S.R.*, **68**, 301 (1949).
[12] Harker, D., *Z. Krist.*, **93**, 136 (1936).
[13] MacGillavry, C. H., and Bijvoet, J. M., *Z. Krist.*, **94**, 231 (1936).
[14] Dunitz, J. D., *Acta Cryst.* (to be published).
[15] Cox, E. G., Sharrett, E., Wardlaw, W., and Webster, K. C., *J. Chem. Soc.*, 129 (1936). Cox, E. G., Shorter, A. J., Wardlaw, W., and Way, W. J., *idem*, 1556 (1937).
[16] Wells, A. F., *J. Chem. Soc.*, 1670 (1947).
[17] Chrobak, L., *Z. Krist.*, **88**, 35 (1934).
[18] Wells, A. F., *J. Chem. Soc.*, 1662 (1947).
[19] Prince, E., and Treuting, P. G., *Acta Cryst.*, **9**, 1025 (1956). See also Bertaut, E. F., *J. Phys. Radium*, **12**, 252 (1951).
[20] Mason, B., *Amer. Mineralogist*, **32**, 426 (1947).
[21] Verwey, E. J. W., and de Boer, J. H., *Rec. trav. chim. Pays-Bas*, **55**, 531 (1936).
[22] Buerger, M. J., *Z. Krist.*, **A95**, 163 (1936).
[23] Hepworth, M. A., Jack, K. H., and Nyholm, R. S., *Nature*, **179**, 211 (1957).
[24] Hepworth, M. A., Jack, K. H., Peacock, R. D., and Westland, G. J., *Acta Cryst.*, **10**, 63 (1957).
[25] Billy, C., and Haendler, H. M., *J. Amer. Chem. Soc.* (in the press).
[26] Stout, J. W., and Reed, S. A., *J. Amer. Chem. Soc.*, **76**, 5279 (1954).

MOLECULAR STRUCTURE AND CARCINOGENIC ACTIVITY OF 1.2 : 8.9 DIBENZACRIDINE

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AGREEMENT between crystallographic determinations and theoretical predictions of bond-lengths in aromatic molecules has not been quantitative, although the usefulness of the fractional bond-order concept has been confirmed. Such discrepancies as do exist can be accounted for in three ways: (1) the bond-lengths as measured in the solid state may not be identical with those for 'free' molecules; (2) the experimental method may be less accurate than previous estimates of accuracy would imply; (3) the theoretical predictions are derived from approximations which may not closely represent the real situation in the molecules, and may not be adequately allowed for in the empirical bond-order – bond-length curve.

If the last of these explanations is responsible for the majority of the supposedly significant discrepancies, any theories of biological activity which relate this activity to particular properties of molecular structure must be open to considerable doubt. Theories of carcinogenic activity in terms of the electron distribution and associated properties of polycyclic aromatic molecules have been summarized recently by Pullman and Pullman[1]. These theories rely on small differences in local electronic distribution to explain considerable variation of physiopathological activity.

To assess the validity of these theories, the crystal and molecular structures of the carcinogenic methyl 1.2-benzanthracenes and the *bis*cyclic dibenzacridines have been investigated. The methyl benzanthracenes, however, do not constitute suitable material for accurate molecular structure analysis and have only been investigated to a stage where the concept of 'incumbrance area' and its relation to activity could be commented upon[2].

The *bis*cyclic dibenzacridines are an interesting group of isomers since, in addition to demonstrating a variation of carcinogenic activity, they form well-defined crystalline complexes with purines and nucleic acids[3]. Accurate molecular structure analysis of the carcinogen and carcinogen–purine complex could therefore be used not only to test the Pullman theory, but also to indicate the nature of the intermolecular forces within the complex. This in turn might throw some light on the mechanism of carcinogenesis.

The preliminary crystal data for the dibenzacridines have already been published[2]. The refinement of 1.2 : 8.9-dibenzacridine is virtually complete (the conventional discrepancy factor for this kind of analysis, $R = \frac{\Sigma| \, |F_o| - |F_c| \, |}{\Sigma \, |F_o|}$, has been reduced to 3·5 per cent) and we may compare the molecular dimensions thus determined with the theoretical model which Dr. H. O. Pritchard and I have calculated, the full details of which will be published later.

Experimental values for the chemically identical but crystallographically non-equivalent bonds of the molecule are given in the first column of Table 1. Usual statistical methods suggest an average probable error of 0·010 A. for these lengths (average estimated standard deviation 0·003 A.). For two chemically equivalent bonds, the maximum deviation from the mean value is 0·009 A., the root mean square deviation being 0·005 A.

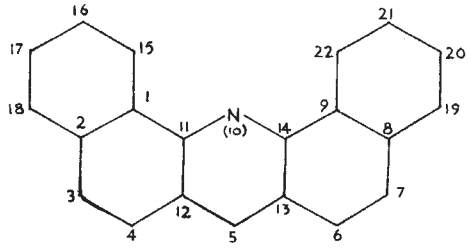

Fig. 1

π-Bond-orders have been calculated by the simple linear combination of atomic orbital – molecular orbital approximation with parameters $(\alpha + 0{\cdot}5\ \beta)$ and $(\alpha + 0{\cdot}1\ \beta)$ for nitrogen and carbon[1], respectively. Two procedures for converting these bond-orders into theoretical bond-lengths will be discussed.

(1) The lengths may be obtained by using the equation

$$R_{ij} = S - \frac{S - D}{1 + \varkappa\left(\frac{1 - O_{ij}}{O_{ij}}\right)} \qquad (1)$$

where S and D are the single- and double-bond distances, respectively, between carbon atoms in an sp^2 state of hybridization and O_{ij} is the mobile bond-order for the bond between any two atoms i and j. Evidence from C—H bond-lengths with carbon in various states of hybridization suggests the values of 1·510 A. and 1·353 A. for S and D, respectively; the best value of $\varkappa$ which will minimize the deviations between the experimental and calculated lengths for this investigation is 0·85. The root mean square deviation of the calculated lengths (Table 1, A) from the mean observed lengths is $0{\cdot}007_6$ A. This is fairly satisfactory, but several individual differences exist which in a statistical sense are significantly larger than experimental errors can account for. This applies particularly to the bonds C(3)—C(4) and C(4)—C(12), for which the deviations between experiment and theory are 0·012 A. and 0·014 A., respectively. It is clear that these bond-lengths are not adequately predicted by the simple theoretical method if equation (1) constitutes a wholly satisfactory method of deriving bond-lengths from bond-orders.

(2) Bond-lengths may also be derived from the bond-orders by evaluating a mean experimental bond-order – bond-length relation. The total bond-orders (the σ-bond-order is assumed to be constant and equal to unity) of all the C—C bonds are plotted against the average experimental lengths and the order–length relation for this particular molecule is derived by finding the best curve through these points. This procedure is legitimate, because there are only three theoretical parameters, S, D and $\varkappa$, to be derived from twelve experimental observations. This experimental curve (Fig. 2) predicts a bond-length of 1·393 A. for benzene as compared with 1·392 A. determined by X-ray crystallographic measurements[5] and 1·397 A. measured spectroscopically[6]; graphite has a predicted carbon–carbon distance of 1·425 A. (experiment 1·421 A.)[7]. The root mean square deviation of the calculated lengths from the mean experimental lengths in 1.2 : 8.9-dibenzacridine is now 0·004 A., and there are no significant differences between theoretical and experimental lengths for any bond in the molecule.

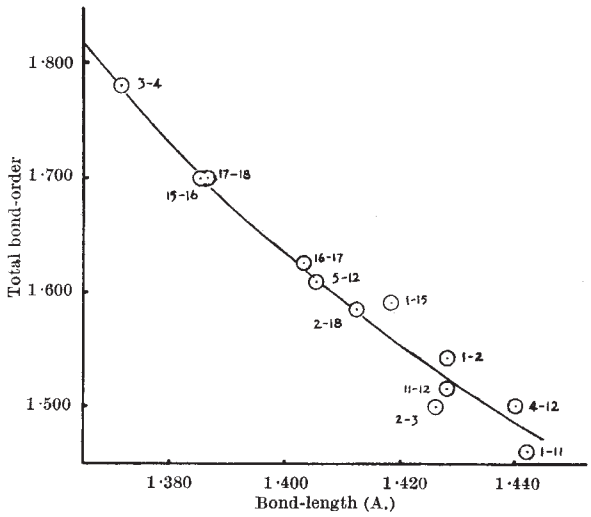

Fig. 2

Table 1

Bond (see Fig. 1)	Experimental bond-lengths (A.)		Calculated bond-lengths (A.)	
		Unweighted mean	A (from equation 1)	B (from experimental bond-order–bond-length curve)
11–N	1·362 and 1·355	1·358	1·358	1·358
1–11	1·449 and 1·435	1·442	1·432	1·446
1–15	1·418 and 1·418	1·418	1·411	1·410
15–16	1·377 and 1·393	1·385	1·395	1·386
16–17	1·399 and 1·406	1·403	1·406	1·402
17–18	1·378 and 1·394	1·386	1·394	1·386
18–2	1·403 and 1·421	1·412	1·413	1·412
2–3	1·423 and 1·428	1·426	1·425	1·435
3–4	1·369 and 1·373	1·371	1·383	1·371
4–12	1·439 and 1·441	1·440	1·426	1·436
12–5	1·399 and 1·410	1·405	1·408	1·406
1–2	1·427 and 1·429	1·428	1·419	1·423
11–12	1·423 and 1·433	1·428	1·428	1·430

The present results therefore suggest that choice of an appropriate bond-order – bond-length relation is a matter of importance, which is contrary to the conclusions reached by Pritchard and Sumner in their investigations of the bond-lengths in anthracene and naphthalene[4]. A re-investigation of the molecular structure of anthracene by Cruickshank using the original three-dimensional data has recently been completed[8]; Dr. H. J. Grenville-Wells and I, using new experimental data, have shown that the more sophisticated modifications of the molecular orbital method which Pritchard and Sumner suggested for anthracene agree well with our present experimental values, but our structure refinement is not yet complete.

Within the experimental errors of the investigation, the molecule of 1.2 : 8.9-dibenzacridine is planar, and chemically equivalent bonds are indistinguishable. Our results suggest that the molecules we have investigated retain their chemical symmetry in the crystal lattice, which, when considered with results such as those mentioned earlier for benzene, indicate that bond-lengths measured accurately by X-ray diffraction techniques are identical, within experimental error, with those measured in the vapour phase. It is only when major steric hindrance effects determine the molecular packing in the crystal lattice that this may not apply.

It can be concluded that bond-orders calculated by simple molecular orbital approximations will give

bond-lengths which are correct to 0·01 A. More detailed theoretical calculations such as those for naphthalene and anthracene may be needed before small differences of electronic distribution within the molecules can be accepted as an explanation of variations of biological activity.

These investigations are being carried out during the tenure of a research grant of the British Empire Cancer Campaign.

[1] Pullman, A., and Pullman, B., "Cancérisaton par les substances chimiques et structure moléculaire" (Masson, Paris, 1955).
[2] Mason, R., *Naturwiss*, **11**, 252 (1956).
[3] Booth, J., and Boyland, E., *Biochim. Biophys. Acta*, **12**, 75 (1953).
[4] Pritchard, H. O., and Sumner, F. H., *Trans. Farad. Soc.*, **51**, 457 (1955).
[5] Cox, E. G., Cruickshank, D. W. J., and Smith, J. A. S., *Nature*, **175**, 766 (1955).
[6] Stoicheff, B. P., *Can. J. Phys.*, **32**, 339 (1954).
[7] Nelson, J. B., and Riley, D. P., *Proc. Phys. Soc.*, **57**, 477 (1945).
[8] Cruickshank, D. W. J., *Acta Cryst.*, **9**, 915 (1956).

DISTRIBUTION OF ELECTROPHORETICALLY DIFFERENT HÆMOGLOBINS AMONG CATTLE BREEDS OF GREAT BRITAIN

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ELECTROPHORETICALLY distinct hæmoglobins have been found among human beings[1] and sheep[2]. Using paper electrophoresis, Cabannes and Serain[3] have shown that adult bovine hæmoglobin exists in two types among Algerian hill cattle. The present article offers data to show the mode of inheritance, and the distribution among different breeds, of the factors controlling the presence of two types of adult bovine hæmoglobin.

The genetical studies were made on the data from 150 matings of three local herds in which both hæmoglobins occurred. Information on the breed distribution of the hæmoglobin types was obtained from 270 stud bulls maintained at the Milk Marketing Board Artificial Insemination Centres in England and Wales.

Electrophoresis separations were carried out on horizontal strips of Whatman 531 filter paper soaked in barbitone buffer at *p*H 8·6 and ionic strength 0·05. Drops of size 1–5 microlitres of an approximately 10 per cent solution of hæmoglobin were put on the paper with a wire loop. Runs were made in an ambient temperature of 5° C. with a d.c. potential of 10 volts per centimetre applied for 18 hr. The papers were stained for two minutes with 2 per cent light green (Messrs. George T. Gurr, Ltd.) in 3 per cent trichloracetic acid and washed to a white background in 4 per cent acetic acid.

In relation to each other the two hæmoglobins are here called Bov. *A* and Bov. *B*. This nomenclature is intended to anticipate the description of yet other electrophoretically distinct hæmoglobins in Bovidæ and yet avoid confusion with the established alphabetic series for human hæmoglobins. The mobility of the Bov. *B* hæmoglobin was greater than that of the Bov. *A*, which, in turn, was slightly less than human type *A*. These results accord with those of Cabannes and Serain[3].

A preliminary survey showed that the hæmoglobin type Bov. *B* could be confused electrophoretically with a type found in new-born Shorthorn calves. This latter type was thought to be fœtal hæmoglobin since it was observed to disappear in from 50 to 70 days, coinciding with the loss of a very rapidly denatured hæmoglobin fraction as tested with *N*/10 sodium hydroxide. In order to avoid any possible confusion, therefore, only animals more than three months old were included in the genetical data.

Table 1. CROSS AND THEIR RECIPROCALS: SIRE, DAM AND DAUGHTERS IN JERSEY AND GUERNSEY HERD

Genotype Parents	Offspring Bov. *A*/Bov. *A*	Bov. *A*/Bov. *B*	Bov. *B*/Bov. *B*
Bov. *A*/Bov. *A* × Bov. *A*/Bov. *A*	1		
Bov. *A*/Bov. *A* × Bov. *A*/Bov. *B*	15	13	
Bov. *A*/Bov. *B* × Bov. *A*/Bov. *B*	3	22	5
Bov. *A*/Bov. *A* × Bov. *B*/Bov. *B*		38	
Bov. *A*/Bov. *B* × Bov. *B*/Bov. *B*		23	23
Bov. *B*/Bov. *B* × Bov. *B*/Bov. *B*			7

Table 1 describes the types of hæmoglobin in numbers of matings from one Guernsey and five Jersey herds. It will be seen that a calf did not possess a hæmoglobin lacking in its parents, and that each hæmoglobin could exist in the presence of the other. From this it is inferred that the different hæmoglobins of these animals could be controlled by a pair of allelomorphic genes, neither of which shows either dominance or recessivity and both being readily recognizable in the heterozygote.

Table 2 gives the type of hæmoglobin found in 271 stud bulls in artificial insemination centres of common breeds of the British Isles. It will be seen that hæmoglobin type Bov. *B* was found only in the Jersey, Guernsey and South Devon breeds. Though a rather small number of animals were examined, it should be pointed out that each animal is contributing one or other gene to a thousand or so head of cattle per annum[5].

Table 2

Phenotype	Nos. with hæmoglobin type Bov. *B*/Bov. *B*	Bov. *A*/Bov. *B*	Bov. *A*/Bov. *A*
Shorthorn			33
Ayrshire			38
Friesian			35
Hereford			28
Welsh Black			7
Aberdeen Angus			13
North Devon			19
Red Poll			5
Sussex			4
Dexter			6
Galloway			1
Jersey	4	22	10
Guernsey	2	10	21
South Devon	1	5	7
Total	7	37	227