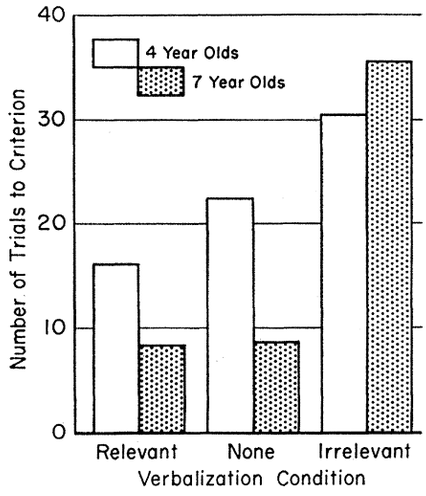

Fig. 2. Effect of verbalization on the acquisition of a reversal shift for 4- and 7-year-old children.

nation to a criterion of nine out of ten successive correct responses, after which the second discrimination was immediately presented without any further instruction or ostensible break in the procedure.

Previous research has found that when no verbalization is required, young children perform a reversal shift more slowly than older children (*2*). This finding is confirmed by the difference between the 4- and 7-year-old, no-verbalization groups of the present study ($p < .05$). To account for these developmental differences it has been suggested that older children are more likely than younger children to make covert mediating responses that facilitate such shifts (*1, 2*).

One likely mediating mechanism is language. The results shown in Fig. 2 lend considerable support to this hypothesis. For both ages combined, relevant labels facilitated and irrelevant labels retarded the shift. These differences, as assessed by analysis of variance, were highly significant ($p < .005$). Apparently, spoken labels that refer to conceptual dimensions *can* mediate a reversal shift in children.

Having demonstrated that their own spoken words are an effective source of stimulation for children and that the experimental procedure employed is sensitive to this effect, it is appropriate to raise questions about possible differences in responsiveness associated with age. Luria (*3*), a Soviet psychologist, has proposed that in the early stages of child development speech is merely a means of communication; not until about 5 years of age does it become a regulator of actions. According to his thesis the effects of verbalization in our experiment should be different for the two age levels. While the results in Fig. 2 suggest that they are different, the implied statistical interaction falls short of significance ($.05 < p < .10$). Consequently, definitive conclusions must await further research. There are, however, some interesting relationships suggested, if not confirmed by these data. For the 7-year-olds, relevant verbalization was no better than no verbalization. This result is consistent with the hypothesis that at this age level children are likely spontaneously to supply relevant mediators and, therefore, need no help from the outside. For the 4-year-olds, relevant verbalization did show a positive influence. This influence was so small, however, that it only decreased slightly rather than eliminated the difference between the ages.

Irrelevant verbalization interfered with the performance of both age groups. The effect for the younger children was again in the right direction but rather weak. For the older children the interference due to irrelevant verbalization was potent, as would be expected if they were particularly sensitive to their own words.

In general, the analysis by age indicates that, at both age levels, the behavior of the children was regulated by their verbalizations. Although no statistically significant evidence indicated that the extent of the regulation was different for the two age groups, this possibility must still be considered (*4*).

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Electron Diffraction from Coals

Abstract. Electron diffraction patterns have been obtained for coals of different rank by transmission through ultrathin sections 500 to 2000 angstroms thick. Analysis of these patterns for the distribution of atoms in coals by Debye radial distribution functions should furnish information complementary to that derived from x-ray studies, considering the differences in wavelength of the radiation involved and the different mechanisms governing the diffraction.

Because fast electrons have much shorter wavelengths than x-rays and their diffraction is proportional to the potential within the crystal (or molecule) rather than to the electron density of the atoms, electron diffraction is a powerful tool in solid-state studies. Heretofore, attempts to obtain electron diffraction patterns of coal have not been successful. Mackowsky and Nemetschek (*1*) reported that finely divided coal preparations graphitized upon exposure to an electron beam. Westrik (*2*) reported a diffraction pattern from a bituminous coal showing a distinct hexagonal crystallinity; the *a* spacing of the hexagonal lattice was 5.2 A compared with 2.46 A of graphite. The pattern was most likely produced by impurities either already present in the coal or introduced during preparation (*3*).

Electron diffraction patterns can be obtained by reflection from the surface of solid specimens. The reflection pattern of an anthracite is shown in Fig. 1*a*. The pattern reveals clearly the usual (002), (10), and (11) (*4*) reflections of aromatic molecules as shown in the densitometer tracing (Fig. 1*b*). Electrons do not penetrate more than several hundred angstroms, and the surface of the specimen is not uniform on the scale of depth of penetration; therefore, the electrons pass through excrescences on the surface. The process has much more the character of transmission than reflection. However, in this method the geometry of the scattering portion of the sample is undefined, and the distance from specimen to plate is uncertain because the diffraction patterns obtained are limited to scattering angles of a few degrees. In addition to these uncertainties, obtaining diffraction patterns from coal surfaces is extremely difficult —that is, it is a hit-or-miss process.

Well-defined electron diffraction patterns can be obtained by using uniform sections sufficiently thin to permit trans-

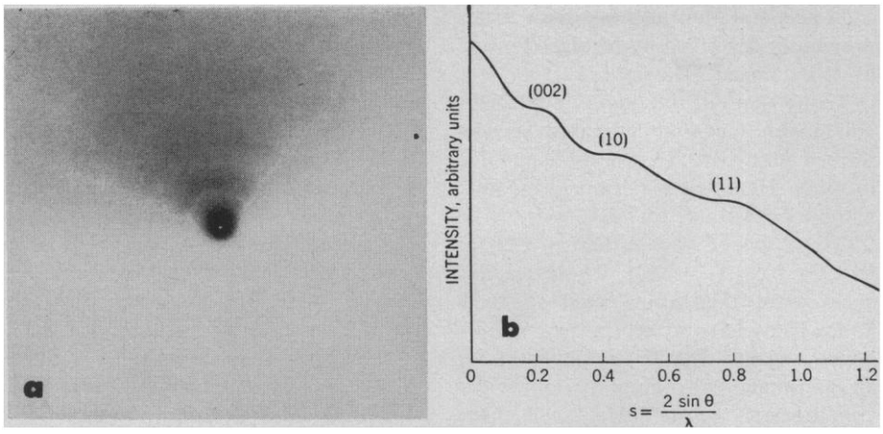

Fig. 1. (*a*) Reflection electron diffraction pattern of an anthracite. (*b*) Densitometer tracing of the same pattern.

mission of electrons. The difficulty in obtaining such sections is probably the reason coals have not heretofore been studied by electron diffraction. With the development of techniques of obtaining ultrathin sections of coals of various ranks (*5*), it is now possible to make such studies. These sections vary in thickness from 500 to 2000 A. Difficulties previously encountered with granular samples are avoided with ultrathin sections. Impurities are not introduced as in grinding, and the ultrathin sections do not absorb enough energy to cause changes. Preliminary qualitative results are reported here. In Fig. 2 are shown the diffraction patterns of vitrinite components of a high volatile A bituminous coal, an anthracite, and a meta-anthracite. The pattern of the high volatile A bituminous coal reveals only the (002), (10), and (11) reflections clearly. The pattern of the anthracite reveals the (002), (10), (004), (11), (006), (20), (21), (30), and (22) reflections of stacked aromatic molecules (indexing was done by examining the negative). The rings belonging to the (002) and (004) reflections, the first and third (very diffuse) rings from the central spot, are uneven in darkening, that is, more or less crescent shaped, indicating an anisotropic structure and preferential orientation in the sample. This is in accordance with observations made of x-ray reflections from slab specimens cut from an anthracite block. The meta-anthracite pattern shows sharp rings corresponding to the three-dimensional reflections of graphite. The following reflections are present: (002), (100), (101), (004), (103), (110), (112), (201), (114), (121), (300), and (220). These peaks are the same as those seen in an x-ray diffraction pattern of this meta-anthracite.

Because the relation between the radial distribution functions of the potential and those of the total charge (atom) distribution is established (*6*), electron diffraction provides two bases for the Fourier synthesis. For noncrystalline materials the synthesis desired most is that for the distribution of the atoms. Representing the distribution of atoms about any one atom by a density function $\rho(r)$ such that $4\pi r^2\rho(r)\,dr$ is the number of atoms between r and $r + dr$ from the atom under consideration, the intensity $i(s)$ in atomic units can be expressed as

$$si(s) = 2\int_0^\infty r\rho(r)\sin(2\pi sr)\,dr \quad (1)$$

where s is defined from $s = (2\sin\theta)/\lambda$, θ being one-half of the scattering angle and λ the wavelength. Applying the Fourier inversion

$$r\rho(r) = 2\int_0^\infty si(s)\sin(2\pi sr)\,ds \quad (2)$$

The experimentally determined values of $i(s)$ are due to the deviation of ρ from ρ_0, the average density of the sample; therefore Eq. 2 should be modified as

$$4\pi r^2[\rho(r) - \rho_0] = 8\pi r\int_0^\infty si(s)\sin(2\pi sr)\,ds \quad (3)$$

Equation 3 can be evaluated numerically by the use of a computer provided $i(s)$ can be obtained from the experimentally observed scattering intensities and provided these intensities extend to a large value of s beyond which $i(s)$ essentially is zero. The latter requirement is the result of the integral calling for $i(s)$ values extending to $s = \infty$. In the case of x-rays $i(s)$ can be calculated directly from the observed intensities, but the range of s is limited; $s_{max} = 1.28$ A^{-1} for CuKα radiation and 2.79 A^{-1} for MoKα radiation. For electrons, however, $i(s)$ is generally determined indirectly and thus is subject to uncertainties, but owing to the shorter wavelength of electrons, scattered intensities extend to much larger values of s, 4 A^{-1} or larger. Because of the uncertainties, quantitative interpretations of the electron diffraction patterns are being approached cautiously. Preliminary to meaningful diffraction analyses, $i(s)$ values obtained from electron and x-ray patterns will be compared in the region of s where they overlap. If these results can be reconciled satisfactorily, it is believed that information gained from the extended range of electron diffraction will be an important complement to that derived from x-ray studies.

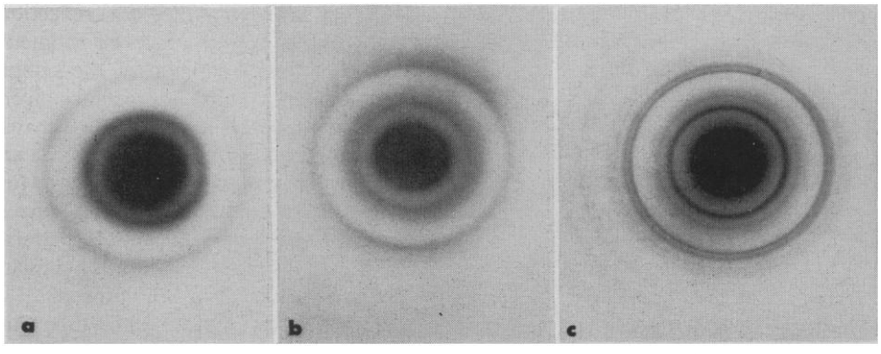

Fig. 2. Electron diffraction pattern from ultrathin sections of (*a*) a high volatile A bituminous coal, (*b*) an anthracite, (*c*) a meta-anthracite.

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24 May 1961

Accumulation of Potassium Anaerobically by Renal Medullary Slices

Abstract. Potassium accumulation occurred in leached slices of the inner medulla of the dog kidney incubated under anaerobic conditions at 37°C with glucose as substrate, but was blocked by inhibitors of glycolysis. Aerobically, only slight accumulation occurred with succinate as substrate at 37°C, and this was blocked by dinitrophenol. These findings were in contrast to those obtained with kidney cortex slices under the same conditions.

Recent work from this laboratory (*1*) showed that tissue slices of the inner zone of the medulla of the dog kidney have a high rate of anaerobic glycolysis and a low rate of oxygen consumption when compared to slices of the cortex of the kidney. It was further found that increasing the osmolality of the medium by addition of sodium chloride caused no diminution in the rate of glycolysis of the medullary slices until the osmolality exceeded 1100 to 1300 mosm/kg of water; the cortical slices, on the other hand, were inhibited progressively as osmolality was increased above the control level of 300 mosm/kg of water. Respiration was similarly inhibited in both types of slices by increasing osmolality.

These findings, considered with other evidence derived from physiological investigations, suggest that the renal medulla *in situ* derives its energy in a milieu of high osmolality mainly from anaerobic pathways. In view of the role of the loops of Henle and the collecting ducts in the countercurrent mechanism for urinary concentration, we have conducted studies concerned with the identification and characterization of active transport systems for ions in this tissue. In the course of these studies, a novel type of potassium accumulation has been found, an accumulation that appears to be dependent upon anaerobic rather than aerobic metabolism. This report is based on studies of this system and compares it to the system for potassium accumulation in the kidney cortex previously described by Mudge (*2*) and by Whittam and Davies (*3*).

Tissue slices of the dog kidney cortex and inner medulla, prepared as described elsewhere (*1*), were incubated in Krebs-Henseleit bicarbonate medium without substrate for 30 minutes at 0°C and gassed with either N_2-CO_2 or O_2-CO_2 (see Table 1). These leached slices were then transferred to fresh Krebs-Henseleit bicarbonate medium containing substrate and reincubated for an additional 45-minute period under conditions shown in Table 1. Then the slices were removed, blotted, weighed, and dried. Potassium was measured on nitric acid digests of the tissue and on the medium by internal standard flame photometry.

Table 1 shows that leached slices of the inner medulla reaccumulate potassium when incubated under anaerobic conditions (at 37°C) with glucose as substrate. This reaccumulation of potassium, although it never reached in vivo levels, was blocked by iodoacetate, fluoride, or ouabain, or by carrying out the incubation at 0°C. Pitressin had no effect. Increasing the osmolality to 1.02 or to 2.10 osm/kg of water by addition of sodium chloride blocked reaccumulation. This effect is probably not mediated specifically through failure of energy metabolism, since previous studies (*1*) have shown that osmolalities in the range of 1.0 osm/kg of water do not inhibit anaerobic glycolysis in medullary slices.

When leached slices of the inner medulla were incubated aerobically with succinate as substrate they showed only slight accumulation of potassium compared with that observed anaerobically. Further losses of potassium occurred from leached slices incubated at 0°C, at 37°C, in the presence of dinitrophenol, or greatly increased osmolality.

Comparative studies of slices of the kidney cortex showed a different pattern of behavior from that observed in the inner medullary slices. Anaerobic incubation of leached cortical slices caused a loss of potassium; iodoacetate, fluoride, and high osmolality all increased the loss, while ouabain and Pitressin had no effect beyond that observed for the control. When the incubation was carried out anaerobically at 0°C, the loss was less than that observed at 37°C.

Aerobically the results confirm those previously reported by others (*2,3*). Reaccumulation of potassium occurred in the complete medium at 37°C but not at 0°C. Losses of potassium occurred from leached slices in the presence of dinitrophenol or increased osmolality.

Table 1. Influence of various factors upon potassium reaccumulation in leached slices of inner medulla and cortex of the dog kidney. The slices were leached in Krebs-Henseleit bicarbonate medium without substrate at 0°C for 30 min. Further incubations were then carried out in a complete medium composed of Krebs-Henseleit bicarbonate medium with 10mM glucose (anaerobic incubations) or 10mM succinate (aerobic incubations). The potassium concentration of the complete medium was 6 to 7 meq /lit. Gas phases were 5 percent CO_2 in either 95 percent O_2 or N_2 at 37°C and 2.8 percent CO_2 in either 97.2 percent O_2 or N_2 at 0°C. Inhibitor concentrations were: iodoacetate, $1.7 \times 10^{-3}M$; sodium fluoride, $5 \times 10^{-2}M$; ouabain, $10^{-6}M$; Pitressin (Parke Davis), 0.2 unit /ml; dinitrophenol, $2 \times 10^{-4}M$; osmolality of the medium was increased by addition of sodium chloride. The concentration gradient for potassium was computed as follows: (μmole K^+/g of tissue water) ÷ (μmole K^+/ml of medium). No appreciable change in the pattern of the data results if the contribution of the extracellular space is neglected in this calculation. The gradient for fresh tissue was calculated by using the determined values for serum potassium. Data shown represent mean and standard deviation; the numbers in parentheses represent the number of experiments.

Conditions	Concentration gradient for K^+	
	Inner medulla	Cortex
Fresh tissue	9.91 ± 0.35 (21)	17.50 ± 0.40 (15)
After leaching at 0°C, gassed with N_2–CO_2	2.21 ± 0.10 (11)	6.15 ± 0.59 (11)
Further anaerobic incubations:		
At 37°C	3.66 ± 0.13 (9)	2.67 ± 0.05 (12)
At 0°C	1.73 ± 0.13 (6)	5.57 ± 0.37 (4)
With iodoacetate	1.42 ± 0.03 (3)	1.71 ± 0.04 (3)
With fluoride	2.25 ± 0.02 (3)	1.88 ± 0.05 (3)
With ouabain	1.91 ± 0.04 (3)	2.83 ± 0.07 (2)
With Pitressin	3.86 ± 0.10 (3)	2.70 ± 0.09 (3)
At 1.02 osmolal	1.38 ± 0.07 (3)	2.23 ± 0.14 (2)
At 2.10 osmolal	1.38 ± 0.06 (3)	1.43 ± 0.06 (3)
After leaching at 0°C, gassed with O_2–CO_2	2.22 ± 0.03 (2)	6.25 ± 0.55 (2)
Further aerobic incubations:		
At 37°C	2.63 ± 0.10 (2)	14.10 ± 0.70 (2)
At 0°C	1.98 ± 0.17 (2)	4.00 ± 0.14 (2)
With dinitrophenol	1.65 ± 0.14 (2)	2.93 ± 0.12 (2)
At 1.04 osmolal	2.28 ± 0.07 (2)	2.19 ± 0.18 (2)
At 1.68 osmolal	1.20 ± 0.01 (2)	1.49 ± 0.02 (2)