With these thoughts in mind, the electron levels of chlorpromazine have been calculated by means of the LCAO method. The K values obtained were -0.217 for the highest filled, and -1.000 for the lowest empty orbital (*4*). This finding is rather striking and unexpected. To our knowledge, chlorpromazine is the first substance ever found in which, in its ordinary stable state, the highest filled level corresponds to an antibonding orbital, as indicated by the minus sign of K. This makes chlorpromazine a quite extraordinarily strong electron donor. Negative values for highest orbitals were hitherto found only in substances on which H atoms were forced by reduction of the molecule in question, as in leucomethylene blue (MBLH) and reduced flavin mononucleotide ($FMNH_2$) (*5*).

In phenothiazine the highest filled molecular orbital also has a negative value and is antibonding (K of the highest filled orbital is -0.210 and K of the lowest empty orbital is -1.000). So, evidently, the strong donor properties of chlorpromazine are linked to the phenothiazine part of the chlorpromazine.

The assumption that charge transfer may be involved in pharmacological reactions of chlorpromazine is supported by the fact that we found *d*-lysergic acid diethylamide, another drug with strong action on the central nervous functions, to be a very good donor. The K value of the highest filled orbital was found to be 0.218; of the lowest empty orbital, -0.726. Also, serotonin is a very good donor, more so than indole or tryptophan (K of the highest filled electronic orbital, 0.461; of the lowest empty orbital, -0.870), and it also has a strong action on the central nervous system. (This agreed with absorption measurements of the complexes (*6*) of these substances, which also indicate that serotonin is a good donor, better than tryptophan.)

The calculations are supported by the chemical behavior of the substances in question. The negative values for MBLH and $FMNH_2$, as pointed out by B. Pullman and A. Pullman, is supported by the great instability of these substances, which readily undergo autooxidation in the presence of O_2. Chlorpromazine, which is not readily autooxidizable, at least not in the dark, is thus unique also in being relatively stable in spite of its energy values. However, the strong donating ability of this drug can be demonstrated by freezing an aqueous solution of chlorpromazine in Dry Ice in the presence of an equimolar riboflavin solution ($10^{-3}M$). On freezing, the solution turns greenish brown and loses all fluorescence—an expression of a complex formation.

Similarly, 1-methyl Medmain is a very good donor (highest filled electronic orbital, 0.348; lowest empty orbital, -0.869). The same is true for 1-benzyl-2-methyl-5-methoxy-(N, N-dimethyl)-tryptamine (highest filled level, 0.427; lowest empty level, -0.866,), which gives strongly colored complexes when frozen with flavin mononucleotide. The assumption that this color is actually due to the formation of a charge-transfer complex is supported by the spectrum. As has been shown earlier (*6*), the spectrum of the serotonin-flavin mononucleotide or of the tryptophan-flavin mononucleotide complex has a peak at 490 mμ, the wavelength characterizing the free radical of flavin mononucleotide at acid *p*H. Also, the chlorpromazine complex, when measured at $-78°C$ (*7*), shows a strong absorption at this wavelength; this indicates that the absorption is actually due to the flavin mononucleotide, which has accepted one electron. In addition, there is a broad absorption at 570 mμ. This peak may be tentatively identified as the absorption of the free radical of flavin mononucleotide at neutral *p*H (*8*). However, other possibilities suggest themselves. The peak may be due to the reverse of the usual charge transfer absorption, in which an electron moves from riboflavin back to chlorpromazine upon the absorption of a light quantum. A positive identification of the 570 mμ absorption must await future work.

If further experience bears out the assumption that the action of the drugs studied, as well as the action of other drugs with related effects, is due to charge transfer, then this may contribute to understanding of the mechanism of normal and abnormal psychic functions (*9*).

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Microbial Fermentation in Certain Mammals

Abstract. The fermentation in the caecum and large intestine of ruminants is negligible compared with that in the rumen. In small ruminants the rate per unit contents is faster than in large ones, due to faster turnover. The cellulolytic bacteria of several ruminants are similar but differ in nutritional requirements.

A sojourn in Kenya to study microbial fermentation rates in the rumen of zebu and European cattle (*1*) also afforded an opportunity (*2*) to examine other mammals. Anaerobic culture tubes containing rumen fluid (or elephant caecal or colonic fluid) and cellulose agar (*3*) were prepared in the field, the contents of the appropriate organ of the freshly killed animal being used as part of the medium. The sterilized tubes were inoculated with serial dilutions of the fresh contents, and the medium was solidified and returned to the laboratory for incubation at 39°C. Characteristic clearing of the cellulose resembling that due to *Bacteroides succinogenes* from cattle (*3*) was observed in cultures from the eland, kongoni, zebu, and camel. A few colonies resembling *Butyrivibrio* (*4*) were also seen. In some culture series, noncellulolytic colonies were sufficiently visible that both total and cellulolytic colony counts could be made. The counts were similar in magnitude to those obtained in cattle by comparable methods (*5*). Data of the experiments are shown in Table 1.

Bacteroides succinogenes-like colonies from the kongoni and the eland, respectively, were inoculated into parallel culture tubes containing media prepared with sterile rumen fluid from the host or from a zebu. Difference in the diameter of the clearings developing in the thin cellulose agar showed that both bacterial strains grew faster with homologous than with zebu rumen fluid, though the difference for the kongoni was slight.

The contribution of each part of the

alimentary tract to the total microbial intestinal fermentation was also measured. The various organs were dissected out and weighed as soon as possible after the animal was killed (Table 2).

Samples of the fresh contents from the rumen, caecum, and colon were transferred as soon as possible to Warburg vessels, and the fermentation was measured manometrically (*1*, *6*). From the fermentation by the sample the total fermentation in each organ and the percentage contribution of each was calculated (Table 3). The rumen fermentation accounted for 95 to 98 percent of the total except in the male camel. Its rumen contained relatively little contents.

The proportion of methane varied between 11 and 24 percent of the total fermentation products. The range for caecal contents was 2 to 30 percent, for the colon, 3 to 26 percent. Its proportion tended to be relatively less in the smaller ruminants (gazelle and suni).

The fermentation rates per unit of rumen dry matter (Table 4) reflect the poor nutrition of the two camels and the zebus of experiment 29. The rate in the suni was the highest observed in any experiments.

Presumably acetic, propionic, and butyric acids were the chief acidic fermentation products, as in domestic ruminants (*7*). The nature of the fermentation products in the suni was of interest because of its very small size and its browsing habit. Direct chromatography (*8*) of 3 g of the suni rumen contents preserved in Bouin's fluid showed both butyric and propionic bands. Acetic acid was presumably present but was masked because the picric acid of the preservative moved in the column at the same rate as acetic acid.

The attempt to measure the fermentation in the elephant caecum and large intestine (experiment 50, Table 1) was unsuccessful, probably because the bicarbonate in the contents had been exhausted. These organs were quite inflated with gas at the time of sampling. Protozoa (thought to be *Elephantophilus zeta*) and numerous roundworms in the caecal contents were visible to the naked eye. The caecal contents were slightly acid to litmus paper; the colon contents were slightly alkaline. The inner surface of the wall of the caecum and about two feet of the colon wall were ridged and folded, presumably as an adaptation for absorption. The stomach contents were distinctly acid. Samples of the caecal and colon contents, removed after fermentation in those organs appeared to have ceased, showed *p*H's of 5.8 and 6.4, respectively, after transport to the laboratory. These values indicate that fermentation stopped before very low *p*H's were reached, as with incubated rumen contents from cattle on a hay ration.

The steady-state concentrations of fermentation products in the caecum and colon of the elephant of experiment 50 were as follows: large intestine, 14 μmole of butyric acid, 23 μmole of propionic acid, 91 μmole of acetic acid, and 6 μmole of formic acid. The kinds and amount of acids are similar to those often found in the bovine rumen.

If the energy requirements of mammals correspond approximately to the ¾ power of the weight, and if in ruminants the rumen fermentation supplies the major part of these requirements, the total fermentation in the smaller ruminants should be greater in proportion to their weight than in the larger ones. An increased total fermen-

Table 1. Culture counts of bacteria in the rumen of various wild and domesticated ruminants and in the caecum and large intestine of the elephant.

Host	Source	Colony count for cellulose digesters (No./ml)	Total colony count, including noncellulolytics (No./ml)
Zebu on grass hay	Ted Ledger, Muguga	2.4×10^8	
Grant's gazelle	Rift Valley, near Muguga	4×10^4	2×10^9
Thompson's gazelle	Rift Valley, near Muguga	2.3×10^6	4×10^8
Eland	Rift Valley, near Muguga	6×10^6	
Suni	Grounds at Muguga		1.3×10^9
Starving Zebu	Archer's Post	6×10^9	6×10^9
Starving Zebu	Archer's Post	4×10^7	
Female camel	Mr. Lowe, Archer's Post	4.1×10^8	
Male camel	Mr. Lowe, Archer's Post	8×10^6	
Fat-tailed hair-sheep	Muguga herd	2×10^6	2×10^8
Elephant caecum	Budongo Forest, Masindi	1×10^5	2×10^9
Elephant colon	Budongo Forest, Masindi	1×10^8	3×10^8

Table 2. Data on weights (in pounds, except when g is used to show grams) of contents in the rumen, caecum, and large intestine.

Animal and wt.	Rumen-reticulum			Caecum		Large intestine	
	Wt.	Ratio to body wt.	Dry wt. (%)	Wt.	Dry wt. (%)	Wt.	Dry wt. (%)
Zebu on fresh grass pasture (530)	80	.15	15.1	3	9.8	2	9.3
Grant's gazelle (108)	10	.09	18.0	68 g	22.9	286 g	17.8
Thompson's gazelle (53)	7	.13	23.9	112 g	18.0	242 g*	19.5
Eland (1143)	122	.11		6	15.1	8	16.1
Suni (3690 g)	290 g	.08	18.7	29 g	15.2	17 g	20.0
Starving Zebu	53		21.1	2	16.4	3	15.3
Starving Zebu	54		14.7	1.5	18.5	2.5	19.4
Camel, very thin, with trypanosomiasis (745)	124	.17	19.6	2	21.2	7.5	25.9
Camel, without food or water for 24 hr (1149)	115	.10	12.4	1.5	17.7	25	18.4
Elephant (3610)		.12†		220*		220*	
Elephant at Paraa (7937)		.11		264		610	
Elephant (2357)		.15		50		300	

* Estimated but not accurately weighed. † For elephants, ratio is of caecum and large intestine contents to body weight.

Table 3. Results of measurements on the fermentation in the rumen, caecum, and colon.

Animal	Fermentation products (mmole/hr)											
	Rumen				Caecum				Large intestine			
	Acid	CO_2	CH_4	% of total	Acid	CO_2	CH_4	% of total	Acid	CO_2	CH_4	% of total
Zebu on grass pasture	526	575	297	95.5	22.6	11.8	3.9	2.6	17.6	8.5	2.2	1.9
Grant's gazelle	70	91	24	97.9	0.67*		0.07	0.4	2.78*		0.34	1.7
Thompson's gazelle	32	60	16.0	97.8	1.14		0.022	1.1	1.23		0.035	1.1
Eland	800	978	488	98.1	20.1		5.3	1.1	15.1		3.45	0.8
Suni	16.2	11.3	3.5	97.4	0.39		0.044	1.4	0.32		.074	1.2
Starving Zebu No. 1	290*		73									
Starving Zebu No. 2	81	148	73									
Camel, very thin, with trypanosomiasis	343	487	164	96.5	7.1		1.7	0.8	23		4.6	2.7
Camel, without food or water for 24 hr	292	109	98	85.5	4.3		1.3	1.0	63		16.0	13.5

* Acid and CO_2 not separately determined.

tation (but slower fermentation per gram of dry matter) could result from retention of food in the rumen for a longer time, permitting more complete digestion. But longer retention necessitates a larger rumen and the rumen/body weight ratios in Table 1 do not indicate a greater ratio with decreasing body size.

The demand for extra energy in small ruminants could also be met by greater consumption of food. Increased food consumption, if not accompanied by an increase in rumen size, would cause faster turnover and a greater fermentation rate per unit of dry matter. The suni, the smallest ruminant studied, showed the greatest fermentation in relation to body weight, the greatest rate per unit of dry matter, and the smallest ratio of rumen contents to body weight. Apparently a greater turnover rate rather than a larger rumen volume accounts for the relatively higher yield of fermentation products. A greater digestibility of the food consumed could also be a factor. Observation of the suni rumen contents disclosed green seeds and foliage rather than grass.

A faster turnover in the small ruminants is also indicated by the lower percentage of methane in the total rumen fermentation products. In hay-fed zebu and European cattle (*1*) a lower percentage of methane is correlated with a higher fermentation rate and decreased retention time. On the assumption that this relationship also holds in wild ruminants, the low methane values indicate faster turnover.

Since more fermentable food would leave the rumen of animals having a rapid turnover, the fermentation in subsequent parts of the alimentary tract should be higher if the turnover rate is the only difference. The rate in the caecum and intestinal contents of the smaller animals is not higher (Table 4), suggesting that factors in addition to turnover affect the completeness of food utilization in the rumen.

From the relationships disclosed in these comparative studies it would be expected that in domestic ruminants with extra energy requirements, as in pregnancy and lactation, an increase in rumen turnover would be the chief means for satisfying the increased need. This would involve increased consumption, presumably through interaction with physiological and psychological factors which control appetite (*9*).

Table 4. Fermentation rates (mmole/hr per gram dry wt.).

Animal	Rumen	Caecum	Large intestine
Zebu	125	37	45
Grant's gazelle	233	45	58
Thompson's gazelle	142	42	42
Eland		53	32
Suni	629	79	64
Zebu No. 1	91		
Zebu No. 2	93		
Female camel	92	46	32
Male camel	59	69	56

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Catalepsy in Two Common Marine Animals

Abstract. Two common marine animals of the South Atlantic and Gulf coasts, *Sicyonia brevirostris*, a penaeid shrimp, and *Opsanus beta*, a toadfish, may be induced to go into cataleptic states in the laboratory when they are threatened or molested. The behaviors of these two animals as they go into catalepsy are described. There is considerable paling in the fish but none in the shrimp.

Catalepsy is an interesting phenomenon which, aside from various fundamental physiological ramifications, may be of interest to surgeons and psychologists in the future in connection with anesthesia, hypnotism, and other matters. It may be defined as a state of bodily inactivity and rigidity in which the organism has an appearance very similar to that of a dead individual.

We have recently observed the cataleptic state in two common marine organisms. The observations seem to be of some intrinsic interest, besides possibly providing a basis for further experimentation.

The first case pertains to a fairly common shrimp, *Sicyonia brevirostris*, one of the so called "rock shrimp" of the family Penaeidae. This is a beautifully colored shrimp, basically white or cream-colored with splotches of pink and old rose on the legs and extremities and purple along the back. It belongs to the family which supports the largest American shrimp fishery, and it is to be found in offshore waters of the Atlantic and Gulf coasts of the United States and Mexico, from Virginia to Yucatan, out to depths of about 40 fathoms. This shrimp has a hard shell and is quite hardy, if kept in water of high salinity. There are other closely related species.

We have noted that when this shrimp is chased about an aquarium and caught by hand, it will go into a rigid state with the body arched concavely from the head to tail and with the legs and swimmerets drawn up tightly against the body. When released, the animal will fall through the water and come to rest on its side on the bottom, where it will lie for several seconds, up to a half minute. No preliminaries to this state have been observed, aside from the usual tail flipping which shrimp display when molested. This state is entered into very quickly, and probably no more than a second intervenes between the active state and the rigid one with the arched back, which is quite characteristic. No color change was noted.

A somewhat different behavior is presented by the toadfish *Opsanus beta*, of the Gulf coast of the United States. This fish is a cognate of *Opsanus tau* of the Atlantic coast, which, because of its large and easily obtainable egg, has become classic material in studies of the embryology of fishes. This species has also been utilized in many physiological studies because it lives in very shallow water and is easily obtainable; furthermore, it is an inactive, bottom fish. The toadfishes have also been studied by students of undersea noises, because of their well-known ability to produce little honking or beeping sounds. The toadfishes have no scales, and their coloring is generally a mottled brown.

We have noted that in the laboratory these fishes may be induced to go into a cataleptic state, but the process is not so simple as that described for the rock shrimp. At first the fish are induced fairly easily to go into catalepsy, but they become more and more recalcitrant as the process is repeated. Initially a fish might go into the cataleptic state when pursued by a few sweeps of the hand around the aquarium. After