trophied fibres due to the greater tetanic contractions accompanied by greater utilization of fat than glycogen.

JOHN[4,5] found that both the specific acetylcholinesterase and the non-specific butyrylcholinesterase are present in the narrow and broad fibres of the pectoralis of *Draco*, and that acetylcholinesterase is slightly higher than butyrylcholinesterase. It was also observed that each fibre contains numerous closely spaced 'en plaque' type nerve endings, suggesting that pectoralis of this lizard is physiologically more active and a fast muscle[9].

Résumé. Le muscle pectoral du *Draco dussumieri* comprend des fibres de trois sortes: minces, intermédiaires et épaisses. Les fibres minces sont adaptées à un métabolisme glycolytique et les fibres épaisses au lipolytique. L'activité des fibres minces révèle la présence de phosphorylase et de synthétase glycogénique.

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Influence of Extra Sucrose, Fats, Protein and of Cyclamate in the Daily Food on the Life-Span of Rats

Studying the life-span of Wistar strain albino rats, it has been found that male animals with 30 Cal/100 Cal sugar in their food lived shorter than controls receiving 14.5 Cal/100 Cal, whereas the life-span of the females was not affected[1]. The study has been repeated with weanling male animals, and groups were added receiving butter, sunflower oil or dried lean meat extra, whereas a 6th group received 0.43/100 g cyclamate. A human type diet was given, composed according to the actual composition data of the Dutch population in 1961, in dried and ground form[2-4]. The variations of this Ran-1961 diet were made by substituting bread and potato starch in proportionate amounts isocalorically by the various compounds. The protein lost by replacing potato and bread was resupplied by cooked potato protein and wheat gluten. Sodium cyclamate was given instead of $^2/_3$ of the sugar in the original ration and in such a way that the sweetening effect was the same, which means $^1/_{30}$ of the weight of the sugar. The sugar content of the Ran-1961 ration is 14.5 Cal/100 Cal. The composition of the rations is given in Table I. The animals were kept – in individual cages – until 'spontaneous' death. Body weights were recorded every 2 weeks. Food and water were always given ad libitum. Kidneys, liver, adrenals, pancreas and testes were examined histologically. Urinary bladders of the control and cyclamate groups have been examined after the first rumours came about cyclamate producing bladder tumors[5-7]. Routine staining was done with haematoxilin-eosin and with sudan (III+IV)-Ehrlich. WILCOXON's test[8] has been used for the statistical evaluation of the data.

Effect of the diets. The animals receiving cyclamate showed a significant higher weight gain and had a better food efficiency than the controls, already in the first weeks. The other groups did not show distinct differences, except for a relative low food consumption and

Table I. Protein and fat content of the diets and fat composition[a]

	I Control Ran-1961	II Sugar extra	III Butter extra	IV Sunflower oil extra	V Dried meat extra	VI Sodium cyclamate food
Protein of dry matter (%)	12.8	12.6	13.7	13.7	23.4	14.7
Fat of dry matter (%)	21.6	21.2	30.6	31.2	23.8	24.9
Fatty acids of total fatty acids (%)						
C12	4.2	4.75	3.6	3.8	4.0	4.35
C 14	6.0	5.6	7.05	4.5	5.2	5.8
C 16	21.5	21.2	22.6	17.8	21.65	21.85
C 16:1	2.5	2.25	2.2	1.75	2.45	2.2
C 17	0.7	0.7	0.75	0.5	0.75	0.7
C 17:1	0.3	0.3	0.35	0.2	0.3	0.3
C 18	12.2	11.95	12.8	9.5	13.15	12.05
C 18:1	29.85	30.55	30.6	28.0	30.0	29.85
C 18:2	14.0	14.35	12.05	27.25	14.4	14.1
C 18:3/20	0.65	0.65	0.55	0.5	0.65	0.65
C 20:1	3.0	2.9	2.65	2.25	2.85	2.9
C 22	0.45	0.35	0.55	0.5	0.35	0.4
C 22:1	1.6	1.45	1.2	1.4	1.5	1.55

[a] Averages of duplicate measurements in each of samples taken with an interval of one year; protein and fat content were controlled in twice as many batches.

Table II. Average weights during the period of study[a]

	I Control	II Sugar extra	III Butter extra	IV Sunflower oil extra	V Dried meat extra	VI Sodium cyclamate extra
Initial (23-day-old)	48.6 (17)	48.5 (17)	48.6 (17)	48.5 (17)	49.1 (16)	48.2 (17)
After: 6 weeks	158.6 (17)	158.5 (17)	147.0 (17)	148.5 (17)	145.3 (16)	188.2 (17)
12 weeks	232.3 (17)	225.6 (17)	206.7 (17)	217.9 (17)	226.1 (15)	264.2 (17)
26 weeks	326.3 (16)	328.0 (16)	322.3 (17)	321.9 (17)	338.2 (14)	343.6 (17)
39 weeks	362.9 (16)	343.5 (16)[b]	342.3 (16)	358.6 (17)	368.5 (13)	369.6 (17)
52 weeks	349.1 (15)	356.1 (13)	328.6 (12)	361.1 (14)	352.8 (16)	350.6 (17)
65 weeks	298.4 (8)	367.9 (8)	328.1 (7)	358.4 (8)	300.7 (3)	309.8 (12)
78 weeks	290.0 (3)	342.6 (5)	344.0 (1)	323.2 (5)		326.2 (6)
91 weeks	365.5 (1)	366.0 (2)		371.5 (1)		221.5 (2)

[a] No. of animals between brackets. [b] Average without the animal that died a few days after weighing the group.

Table III. Maximum weights, age at maximum weight, life-span, body weight at death and weight of organs (averages and ranges)

	I Control	II Sugar extra	III Butter extra	IV Sun-flower oil extra	V[a] Dried meat extra	VI[b] Food with sodium cyclamate	P_2 WILCOXON[c]
Maximum	396	378	373	387	376	395	(I+II+IV+VI)-(III+V): n.s.
weight (g)	249–464	277–517	285–510	226–484	255–461	310–461	
Age at maximum	304	318	290	312	273	325	(I+II+IV)-(III+V): 0.05
weight (days)	163–394	191–646	191–394	205–394	163–380	177–478	(I+II+IV)-VI: n.s.
Life-span (days)[d]	481	472	433	485	405	528	I–III: 0.18; I–V: 0.016; I–VI: 0.12;
	177–815	201–711	289–597	320–677	148–535	415–723	(I+II+IV)-(III+V): 0.004; (I+II+IV)-VI: 0.05
No. dead before	4	6	6	5	10	0	
415 days old							
Body weight at	233	248	241	247	232	241	
death (g)	147–336	183–338	167–368	172–357	175–323	180–388	
Liver weight (g)	16.8	17.1	18.7	16.2	18.6	16.7	
	7.5–23.6	12.4–24.1	11.2–25.7	10.0–24.2	12.1–27.5	8.8–21.4	
Weight of	3.30	3.54	3.20	3.22	4.12	3.30	I–V: 0.01
kidneys (g)	2.00–4.64	2.24–5.21	1.77–4.87	1.94–8.66	2.10–5.70	1.89–4.92	
Weight of	1.20	1.30	1.20	1.32	1.31	1.12	
testes (g)	0.61–2.25	0.57–2.50	0.63–2.08	0.70–2.10	0.61–2.31	0.47–2.49	
Weight of	55.0	58.8	53.1	50.7	59.7	48.3	I–VI: n.s.
adrenals (mg)	35.5–79.0	16.0–87.2	43.9–72.7	35.9–76.5	39.9–93.0	32.8–80.0	
Weight of	0.49	0.48	0.46	0.53	0.44	0.47	
pancreas (g)	0.14–0.84	0.20–0.85	0.17–0.74	0.24–1.04	0.26–0.66	0.15–0.82	

[a] Two animals died at age 51 and 83 days respectively. Their data are discarded. [b] The organ weights of the cyclohexylamine producing animals were not different from those of the other animals. Only 1 produced seizable amounts of cyclohexylamine – at least 5 mg a day –; it survived for 433 days. [c] P_2, bilateral tail error. [d] The second last animals survived for 594, 669, 514, 636, 519 and 641 days respectively.

high food efficiency of those receiving meat extra, although the latter was not as high as in the cyclamate group[2].

Table II gives the initial weights of the animals and weight changes during the study. The cyclamate group kept the highest average weight till about the 80th week, although the differences with the other groups became smaller. Better growth with cyclamate has also been seen by SABRI et al.[9] dependent on the basic food given. The food consumption of the adult animals showed only minor differences between the groups, which were caused mainly by differences in average body weights. The calorie intake in the 40th and 41st weeks per g body weight at week 41 (310-day-old) was in the groups I through VI respectively: 2.40, 2.43, 2.49, 2.59, 2.29 and 2.50 with average weight changes in this period of +4.0,

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+5.5, −1.9, +5.4, −7.0 and +8.1 g. Life-span was shortest in the groups showing early average weight decrease, that is in those receiving butter or meat extra (groups III and V).

The cyclamate group had the longest average life-span, not so much because of animals surviving very long, but owing to a much higher age at which the first death occurred (Table III). The latter might be connected with a weak sulfa-drug like activity of the cyclamate[10]. The average total amount of cyclamate consumed during life is estimated to be about 40 g.

Extra sugar in the food did not shorten life-span as contrasted with the earlier findings. In fact, controls had a shorter life-span than those in the previous experiment. The average age at death was then 486 days in the extra sugar group and 566 days in the control group, whereas in the present study 472 and 481 days were found respectively. An explanation cannot be given, but it is clear that other factors than the sugar consumption alone play a role.

Autopsy. Macroscopically changes were seen – apart from chronic respiratory disease – especially in liver and kidneys. Apart from kidney tumors no tumors were apparent. Some organ weights are given in Table III.

In 55% of all animals the liver showed a superficial necrosis, which only seldom reached into the more central parts. Congestion was frequent in all groups (about 65% of the animals) except for the early dying group with extra meat (V), whereas in the group with extra butter (III) it was seen especially in the longest survivors. Necrosis and fatty degeneration were seen in over 90%; oedematous gall capillaries were found in about 15%.

The kidneys were enlarged to 2–3 times the normal size in almost all animals, were greyish and had pitted surfaces. They were nearly white in 9 of the 15 animals on the extra meat food (excluding the two animals that died in the first experimental weeks). The high kidney weight of this group is also in agreement with a very acute course of the glomerulonephritis in these animals, which was obviously the cause of death.

Microscopically the average glomerulonephritis score – using a 0–4 scale – was 3 and necrotic areas were present in 100%, whereas that in the other groups varied between 1.5 and 2.6 with necrotic areas in 60%. The latter animals were on an average older than the former. Similarly the score for glomerulus sudanophilia – on a 0–3 scale – was 3 against 1.1–1.7 for the others. All had sudanophilia of the hilus vessels, which was true for only about 20% of the animals in the other groups.

Five large kidney tumors (Wilms tumors) were found distributed over the groups I–IV; 2 of a few mm diameter escaped further investigation because they fell out of the slides. There might have been more of the latter, since only 1 kidney per animal was studied. 3 animals had a large haemorrhage/infarction (?) of a kidney pole and 1 (group III) had a thick stratified epithelium covering the papilla up to far into the angle between papilla and cortex resembling the picture preceding formation of kidney stones on high calcium carbonate food[11]. Kidney stones were not found. A few calcified glomeruli were seen in about 8% of the animals.

Testis atrophy was often clearly seen macroscopically; microscopically it was definite in 85%. About 20% showed an orchitis presumably of viral origin of a type found for at least 20 years in this strain of anialsm. Signs of periarteritis were seen in about 30%.

The adrenals showed haemorrhages and necrosis in about 35%; congestion in 25%. Group VI had a tendency to lower adrenal weights than the other groups.

In animals of all ages the pancreas had extremely high necrosis scores – in a scale of 0–4 the average was 2.4. Often no islets of Langerhans were seen, but glucosuria was never found. Periarteritis was found in about 30%. The pancreas changes show much resemblance to those described by KENDREY and ROE[12].

No urinary bladder tumors were found in the cyclamate and control animals that could be studied (about $^1/_2$ of the groups). The daily dose, however, was only about $^1/_{10}$ of the carcinogenic dose of PRICE et al.[7] and moreover no cyclohexylamine was administered in the present study. Only 1 animal (the 3rd one that died) of the group proved to be a reasonably good converter.

There were no differences between the groups except for the more severe kidney changes in the extra meat group than in the other groups[13]. Age trends in the pathological findings were not clear. Even testis atrophy was not obviously more severe in those surviving longest.

In conclusion, it can be said that animals receiving dried meat or butter extra substituted for starch in a human type food lived less long than control animals, animals with extra sugar or animals with extra sunflower oil in their food. Between the latter 3 groups no difference in longevity was found. A 6th group receiving cyclamate instead of $^2/_3$ of the sugar normally present in this food, and given in the appropriate sweetening amount, reached a higher body weight and had a tendency to live longer on an average. Remarkable was that the first animals of this group that died did so in a far later stage of the experiment than the first that died in the other groups.

Regarding earlier findings, it must be concluded that life-span is definitely influenced by other factors than diet alone. Their nature is still unknown, but might be connected with infectious diseases occurring among the animals.

Definite differences in causes of death between the groups were not seen, except for a tendency to earlier and more severe glomerulonephritis in the group receiving extra meat[14].

Résumé. Une quantité supplémentaire de 15 Cal/100 Cal de viande maigre sèchée ou de beurre dans un régime humain typique diminue la durée de vie du rat Wistar par comparaison avec des témoins et des animaux qui avaient reçu dans leur régime une quantité supplémentaire de sucre ou d'huile de tournesol. La durée de vie de ces derniers groupes fut la même. Un autre groupe qui avait reçu dans son régime de cyclamate de sodium au lieu de $^2/_3$ de la quantité normale de sucre présenta un poids plus élevé et une durée de vie plus longue que les autres groupes.

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[13] In animals on stock food – containing about 1.5 as much protein as the Ran food – glomerulonephritis does not occur so early in life, is less severe and less rapidly progressing than in animals on the human type food. Their life-span is appreciably longer than that of the latter, but a protein supplement to this food still does shorten the life-span of the male animals, although not that of the females.

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