approximately 10 cm apart. When this flask is shaken vigorously, electrical breakdown phenomena are seen inside. The experiment must be performed in a dark room and the observer must be well adapted to the dark (a process taking at least 15 minutes). Observers have concurred that several modes of breakdown can be observed: (i) small sparks, (ii) discharges which appear to be several centimeters long and about 1 cm wide—both of these are relatively bright, and (iii) a faint extensive glow discharge. The dominant colors are in the blue to red parts of the spectrum. The sensitivity curves for scotopic vision are known to be shifted toward longer wavelengths relative to photopic vision, with the minimum perceptible brightness of the order of $10^{-4}$ mlam.

When a d-c potential of about 1200 volts was impressed between the two electrodes, similar discharges were produced without the necessity of any dust movement. Some estimate of the electrification occurring on shaking was also obtained. After agitation, it was observed that potentials as high as 500 volts appeared between the two electrodes. During the course of these experiments, the surface of the inside of the flask became coated with fine particles which adhered strongly.

We stress that this experiment is not intended to be an analog of the martian atmosphere; however, the possibility of related effects occurring there warrants consideration. The atmosphere near the surface of Mars is primarily $CO_2$ at a pressure of about 10 mbar. The surface is swept by severe winds and consequent dust storms which occasionally are of global dimensions (*6*) and which may affect the thermal structure of the atmosphere (*7*). Mariner 9 coincided with such a storm. Photoionization, which may contribute to the conductivity of the lower atmosphere on the light side, must be reduced on the dark side, and the conductivity of the lower atmosphere will be primarily due to fast ions caused by cosmic rays or, possibly, by radioactive material in the surface. The rate of production of fast ions may not greatly exceed corresponding values in the earth's atmosphere at about 10 mbar. High conductivity is then unlikely because of the capture of fast ions by dust particles, unless there is a very high anomalous rate of ion production. It seems reasonable to suggest that excitation of the atmosphere with emission in parts of the spectrum other than the visible, or even spark breakdown or glow discharges in the visible, might result as a consequence of the electrification of the dust in such storms.

Dust storms may play a finite but small role in the electrical budget of the atmosphere of the earth, which is primarily the balance between the earth-atmosphere current in fair weather and thunderstorm activity. We are considering what significance dust storms on Mars may have for the electrical budget of that atmosphere both by generating charge and by reducing the conductivity by the capture of fast ions.

Because of dust electrification an engineering problem could occur with a martian lander vehicle. Dust blown by or over such a vehicle may result in charge generation. Helicopters hovering near the surface of the earth can electrify rapidly due to blown dust, and breakdown phenomena occur. Surfaces exposed to moving dust rapidly become coated with fine particles, which adhere by means of Coulomb forces that greatly exceed the gravitational forces on them. Similar effects could occur with a lander vehicle, including the possibility of exposed lenses becoming coated, which would degrade visual data (*8*).

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## Number of Germ Cells in Known Male and Known Female Genotypes of Vertebrate Embryos (Oryzias latipes)

Abstract. *The number of primordial germ cells in embryos of known genotypic sex was the same in XY males and XX females until the gonad became recognizable as testis or ovary. It has been claimed that the heterogametic sex chromosome causes the gonad to differentiate as a testis in mammals and as an ovary in birds as a result of earlier and more mitoses. This claim was not supported in the present study where a sex difference in numbers of germ cells was first noted during differentiation of oocytes in the XX embryos.*

The number of primordial germ cells has been thought to differ with sex (*1*). For example, in embryos of the elasmobranch *Raja batis*, counts were grouped around two numbers (256 and 512) which were claimed to correspond, respectively, to future males and females (*2*). Yet in a recent review of all data for vertebrates, Hardisty concluded that it is not clear whether these sex differences are "due to earlier or more rapid proliferation in the germ line of one sex or to differences in the number of primordial germ cells originally segregated" (*1*). In studies made over almost a century, a major difficulty has always been uncertainty as to the sex of an individual specimen prior to differentiation of the gonads. Of obvious value would be data for species in which the sex of every embryo is known from the time of fertilization.

Table 1. Numbers of germ cells in embryos and hatchlings of known genotypic sex in the d-rr strain of *O. latipes*.

| Stage | Number of embryos/ hatchlings | Mean ± S.E. | Range |
|---|---|---|---|
| 26-1 | 16XX/ | 32 ± 2.1 | 20–47 |
| | 16XY | 34 ± 2.5 | 17–47 |
| 30 | 16XX/ | 32 ± 1.1 | 23–43 |
| | 16XY | 35 ± 2.1 | 23–48 |
| Hatching | 16XX/ | 75 ± 11 | 23–198 |
| | 16XY | 72 ± 6.3 | 28–107 |

Therefore, our study was undertaken with the Japanese killifish, *Oryzias latipes*, in which sex determination is of the XX-XY type, with the female homogametic and the male heterogametic (*3*). Special procedures (*4*) applicable to the d-rr strain, provided fertile XX males and YY males which were bred with XX females to produce zygotes known to be XX and destined to develop as females, or

known to be XY and destined to develop as males.

Four mature females with XX sex chromosomes spawned the eggs studied. These females were bred first with four XX males and subsequently with four YY males. In an initial control experiment, 40 progeny of each mating were raised to sexual maturity, with the result that all of the XX fish were of the female phenotype and all of the XY progeny were of the male phenotype.

Thereafter, embryos of known XX or of known XY genotype were collected at embryonic stage 26-1, at which time the germ cells are migrating to the gonadal primordium from the extraembryonic endoderm; at embryonic stage 30, at which time the primordial germ cells have just completed migration to the gonadal primordium; and at hatching, at which time gonial proliferation is beginning (*5, 6*). At each of these three stages, 16 XX and 16 XY specimens were collected and prepared by Gamo's method (*6*). This technique requires fixation in Bouin's solution, dechorionation of the embryos, embedding in paraffin with a melting point of 53° to 55°C, and serial sectioning at 6 $\mu$m.

The origin of the germ line and differentiation of the gonads has been described in *O. latipes* (*6, 7*), but no use was made of embryos of known genotypic sex. In our work, the first in which germ cells were studied in embryos whose sex was known with certainty, there was no sex difference in the number of primordial germ cells either during migration to the gonadal primordium or after arrival in the primordium (Table 1). At hatching, there was only a slight sex difference in the number of germ cells with the onset of their proliferation. This proliferation culminates within 2 days after hatching in significantly more germ cells in presumptive ovaries than in presumptive testes, as has been reported for the d-rR strain of *O. latipes* (*7, 8*), a time when sex differentiation of germ cells has taken place (*8*). This we confirmed in progeny of known sex genotypes produced by other breeders in the d-rr strain (Table 2).

Earlier and more mitoses in the entire embryonic gonadal rudiment of the heterogametic than of the homogametic sex is currently claimed (*9*) to cause differentiation of the mammalian testis (and of the avian ovary) and to be a function of the "odd" sex chromosome which is the Y in mammals. If this interesting concept applies to a teleost such as *O. latipes* (in which the "odd chromosome" is the Y and is associated with differentiation of a testis), it is not applicable to the number of germ cells proliferated by hatching and shortly thereafter, because the number was not larger in the testis than in the ovary.

In conclusion, the number of primordial germ cells, as originally segregated in *O. latipes*, is the same in homogametic embryos (XX females) and heterogametic embryos (XY males). At a later stage of development, mitosis of these cells within the gonads results in more germ cells in females than in males.

Table 2. Number of germ cells at 2 days after hatching in the d-rr strain of *O. latipes*: Animals of known genotypic sex produced by ten pairs of parents.

| Animals | Germ cells (mean ± S.E.) | Range | |
|---|---|---|---|
| 10XX | 157.4 ± 20 | 81–354 | $P < .001$ |
| 10XY | 76.0 ± 20 | 39–136 | |

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## Isotachysterol$_3$ and 25-Hydroxyisotachysterol$_3$: Analogs of 1,25-Dihydroxyvitamin D$_3$

Abstract. *Isotachysterol$_3$, 25-hydroxyisotachysterol$_3$, and isovitamin D$_3$ have been synthesized and tested for biological activity. Like 1,25-dihydroxyvitamin D$_3$, isotachysterol$_3$ stimulates intestinal calcium transport and bone calcium mobilization in anephric rats, whereas 25-hydroxyvitamin D$_3$ does not. Although isovitamin D$_3$ is biologically active in normal rats it is inactive in anephric rats.*

Vitamin D must first be hydroxylated on carbon-25 (C-25) in the liver (*1, 2*) and then on carbon-1 (C-1) in the kidney (*3*) before it can carry out its physiologic functions. Unlike vitamin D or its 25-hydroxy derivative, 1,25-dihydroxyvitamin D$_3$ (1,25-(OH)$_2$D$_3$) is capable of stimulating both intestinal calcium transport and bone calcium mobilization in anephric rats (*4*). These results make clear the potential use of 1,25-(OH)$_2$D$_3$ in the treatment of patients with hypocalcemia, impaired intestinal calcium transport, secondary hyperparathyroidism, and osteodystrophy due to renal dysfunction. However, be-

Table 1. Intestinal calcium transport and bone calcium mobilization response to isotachysterol$_3$, isovitamin D$_3$, 5,6-*trans*-vitamin D$_3$, 25-hydroxyisotachysterol$_3$, and 25-OH-D$_3$ in normal and anephric rats.

| Compound | Amount ($\mu$g) | Animals (No.) | Ca in serum (mg/100 ml) (mean ± S.E.) | $^{45}$Ca serosal/$^{45}$Ca mucosal (mean ± S.E.) |
|---|---|---|---|---|
| | | *Normal* | | |
| None | 0 | 6 | 4.3 ± 0.1 | 1.8 ± 0.2 |
| Isovitamin D$_3$ | 5 | 6 | 6.5 ± .1 | 3.5 ± .3 |
| Isotachysterol$_3$ | 5 | 6 | 6.3 ± .1 | 3.2 ± .3 |
| 25-Hydroxyisotachysterol$_3$ | 5 | 6 | 6.4 ± .1 | 3.0 ± .2 |
| 25-OH-D$_3$ | 0.25 | 6 | 6.4 ± .1 | 3.2 ± .2 |
| 5,6-*trans*-vitamin D$_3$ | 5 | 6 | 6.0 ± .1 | 3.5 ± .1 |
| | | *Anephric* | | |
| None | 0 | 6 | 3.7 ± 0.1 | 1.2 ± 0.2 |
| Isovitamin D$_3$ | 5 | 6 | 3.8 ± .1 | 1.2 ± .1 |
| Isotachysterol$_3$ | 5 | 6 | 5.0 ± .1 | 3.2 ± .2 |
| 5,6-*trans*-vitamin D$_3$ | 5 | 6 | 4.9 ± .1 | 3.5 ± .2 |
| 25-OH-D$_3$ | 0.25 | 6 | 3.9 ± .1 | 1.4 ± .2 |