from a collection made in 1965 at an isolated single tree 700 metres away from the above locality. 24 males of the next generation were found to be 24 $(22+XY_1)$ and 1 was 24 $(22+XY_2)$. A locality at which 25-chromosome males did not occur was found by Callan⁵ at Merton (England).

Evidence from our second locality and from mosaic individuals indicates that it is the Y_2 chromosome which accumulates in the cells of the line of males in which it occurs. It is not known if it can be transmitted in multiple Y_2 sperm or if it is accumulated in post-zygotic development.

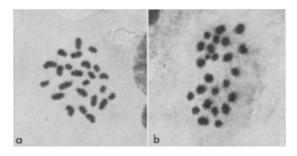


Fig. 1. Chromosomes in C mitosis in ovarian follicle cells of $Forticula\ auricularia$. a) 24 chromosomes; b) 24 chromosomes plus one fragment. Both $\times 1330$.

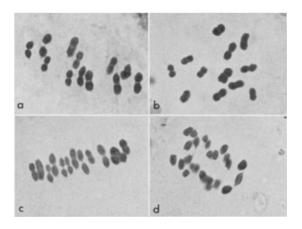


Fig. 2. Meiotic chromosomes of Forficula auricularia males. a) First metaphase, side view, karyotype 24 $(22 + XY_1)$, sex bivalent second from right. b) First metaphase, polar view, karyotype $24(22 + XY_2)$, one chromosome of the sex bivalent bilobed. c) First metaphase, side view, karyotype 25 $(22 + XY_2Y_2)$, sex trivalent second from right. d) Early first anaphase, side view, karyotype 25 $(22 + XY_2Y_2)$, apparent meiotic error with the Y_2 chromosomes migrating to each pole, X chromosome not committed. All \times 1200.

A small but significant excess of males (56.3%) was found in the Melbourne University collection. This fact does not support a hypothesis of Lewis and John⁴ based on X_1X_2Y sex chromosomes in 25-chromosome males. These authors explained the excess of females found in many populations of $Forficula^9$ as being due to meiotic errors producing an excess of female-determining sperm in these males. Meiotic errors were noted in 25-chromosome males of the present study but on the new interpretation they would lead to an excess of Y_2 bearing sperm (Figure 2d).

The apparent necessity for a Y chromosome for male determination in the cytologically known species of Dermaptera 6 has influenced our designation of the Y_2 chromosome. Otherwise it might be considered to be a supernumerary chromosome which segregates from the X during meiosis 10 , 11 .

Henderson¹² has recently published a detailed study of males of a population of *Forficula auricularia* at Cambridge, interpreting 25 chromosome males as XXY. He interpreted variation in the size of the sex chromosomes as a complex polymorphism of both the X and Y. We have made similar observations but interpret the situation as due to increases in the size of some of the Y_2 chromosomes.

Riassunto. Osservazioni su femmine di Forficula auricularia L. indicano che esse possiedono un solo cariotipo formato da 22 autosomi e da 2 cromosomi X. Sulla base di questo studio, suggeriamo che i maschi con 3 cromosomi sessuali sono del tipo XYY, e non XXY come precedentemente ritenuto da varii autori. Ci sono maschi XY_1 , XY_2 , XY_2Y_2 e mosaici XY_2/XY_2Y_2 nella popolazione. Inoltre viene dimostrato che solamente il più grande dei due cromosomi Y, che caratterizzano questa specie, viene accumulato nei maschi con un meccanismo multiplo di cromosomi sessuali.

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Amino Acid Excretion Patterns in the Offspring of a 'Doubly-Heterozygous' Cystine Stone Former

The genes regulating the expression of the different biochemical types of cystinuria may be allelic, as more than one form of the disease has recently been found within the pedigree at 4 such families are on record, 2 carrying Types I and III cystinuria 3, 1 with Types I and III, and 1 with Types II and III. The cystinuria in at least 3 other families, described before the 3 types were defined 4-6, may also be heterogeneous. Stone formers in

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Excretion of cystine and the dibasic amino acids by the 2 daughters
of a Type I-III cystine stone former

Source	mg/day					
	Sample	Cystine	Lysine	Argi- nine	Orni- thine	Homo- cysteine Cysteine
C.	1st 2nd	8 13	37 62	2.9 3.4	1.5 2.5	3.3
E.	1st 2nd	15 13	73 47	3.5 4.4	2.9 2.1	3.9
Normal,	up to	37	28	4.0	2.0	2.3

the families of record were recognized as 'double heterozygotes' through the different excretion patterns of cystine and the dibasic amino acids by 2 or more presumed carriers in each pedigree, either by the 2 parents or by 2 or more children of the stone formers. The 'double heterozygotes' resembled homozygotes clinically and biochemically and were not distinguished without the family studies.

Our analysis of amino acid excretion rates in 2 children of a Type I–III 'doubly heterozygous' stone former 3 adds more data to the evidence for allelism in cystinuria and broadens the interpretation of the carrier state in the Type III form.

Materials and methods. Urine from the 2 children of a Type I-III stone former, both healthy daughters, aged 4 (C) and 7 years (E) when first sampled, was collected for 24 h with HCl preservative and was assayed for amino acids by nitroprusside reaction,7, paper chromatography^{8,9}, and ion exchange chromatography in a Beckman 120C analyzer 10. The daily excretion rates of cystine and the dibasic amino acids were calculated from the column chromatographs and were compared with published values 11 and with rates from 9 children of laboratory personnel, aged 3 to 13 years. The mixed disulfide of cysteine-homocysteine was identified in assays in which the buffer change in the 59 cm column of the analyzer was postponed until 340 min 12. Second samples were obtained from the girls 2 years later and were assayed by similar procedures.

Results. Positive nitroprusside-cyanide reactions and paper chromatography suggested that the first samples of urine from both girls contained excessive concentrations of cystine. The daily excretion rates of cystine, however, were within normal ranges. The excretion rates of lysine and of the mixed disulfide, cysteine-homocysteine, at the first sampling, were greater than normal. Those of ornithine and arginine by the older girl were in the 'high normal' ranges (Table). Methionine sulfoxide excretion by both was normal.

The rates of cystine excretion by the girls when they were 2 years older were again normal. Lysine excretion rates were still greater than normal, however, and those of ornithine and arginine were in the 'high normal' ranges for both (Table).

Discussion. The offspring of either homozygous or 'doubly heterozygous' cystine stone formers are heterozygotes, if the other parent is normal. They may or may not express the gene defect biochemically, depending upon the form of cystinuria of the affected parent. If the genes for the 3 known disease forms are alleles2, the biochemical characteristics of one or another form should be found in the individual offspring, not those of 2 forms combined. The 2 daughters of the Type I-III stone former, a 'double heterozygote', should excrete, accordingly, either normal amounts of the amino acids, as do Type I carriers (and their grandmother), or slight excesses of cystine and the dibasic amino acids, as do Type III carriers (and their grandfather).

We consider both girls heterozygotes of the Type III form of cystinuria, as they excrete excesses of lysine and of the mixed disulfide and 'high normal' amounts of ornithine and arginine. Excessive excretion of cystine is not a sine qua non, apparently, of Type III carriers, as several, excreting excesses of lysine only, have been described in 'incompletely recessive' families or in those with Type III cystinuria 3, 13, 14.

Résumé. L'analyse des acides aminés de la cystinurie du type I-III dans l'urine de deux enfants d'un hétérozygote double a révélé que les quantité d'acides aminés dibasiques excrétées en 24 h sont celles qui furent trouvées chez les hétérozygotes de la cystinurie du type III. Les données expérimentales s'accordent sur ce point que les types biochimiquement hétérogènes de la cystinurie sont règlés par des allèles.

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Replication of Intranucleolar DNA in Smittia (Diptera, Chironomidae)

One of the major functions of nucleolus is the production of ribosomal RNA and the genes which code for this class of RNA are localized in or near the nucleolar organizer in Drosophila1 and Xenopus2. In animal and plant cells, it has been known for a long time that the organizer is the specific site of formation of the nucleolus but various aspects, structural and functional, of the nucleolus-organizer chromatin complex still remain to be

elucidated. In recent years it has come to be widely recognized that the chromatin of the organizer may

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