then the sample (1 vol.) and finally 0.1 vol. of oil to enclose the sample.

We have successfully used this piece of apparatus for the transfer of small volumes of fluid for the estimation of inositol[3] and carnitine[4] in the fluid collected by micropuncture from the seminiferous tubules of the mammalian testis and epididymal duct.

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Ag-G staining, a rapid technique for producing combined silver staining and Giemsa banding in mammalian chromosomes

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Summary. A combination of the silver staining method with a Giemsa banding technique is described for mammalian chromosomes. This double staining simultaneously shows silver-stained NORs and a G-band pattern, and thus allows a rapid identification of the NOR-bearing chromosomes.

Ammoniacal silver (A-S) has been used to stain differentially chromosomes which are involved in the genetic activity of the NORs[1,2]. A number of techniques have been developed for the staining of NORs on human chromosomes[3-6]. These methods have the disadvantage that the chromosomes are only slightly, and uniformly, stained with the exception of the black-stained NORs. For identifying these NOR-bearing chromosomes additional techniques had to be applied. Lately combined methods have been described where silver staining was followed by Giemsa banding[7,8]. These combined procedures are very suitable for human chromosomes but less favourable for other mammalian chromosomes. In this paper we describe such a combined technique and its application to mammals of the family Bovidae.

Materials and methods. Lymphocyte cultures from peripheral blood from 3 individuals of cattle *(Bos taurus)* and goat *(Capra hircus)* were set up after routine methods. Chromosome preparations were stained with a modified silver method according to the Ag-staining of Mikelsaar et al.[9,10]. 4 drops of 50% aqueous $AgNO_3$ were put on air-dried chromosome preparations and covered with a cover-glass. The slide was placed for 10 min under a photoflood light (temperature optimum at 60 °C). After this procedure the slides were rinsed in several changes of distilled water and air dried.

For the next step, 2 drops of solution A (4 g $AgNO_3$ dissolved in 5 ml NH_4OH and 5 ml aqua dest.) and 2 drops of solution B (3,2 g Na-acetate in 97 ml aqua dest. and 3 ml formaldehyde) were mixed on a cover glass, and the slide

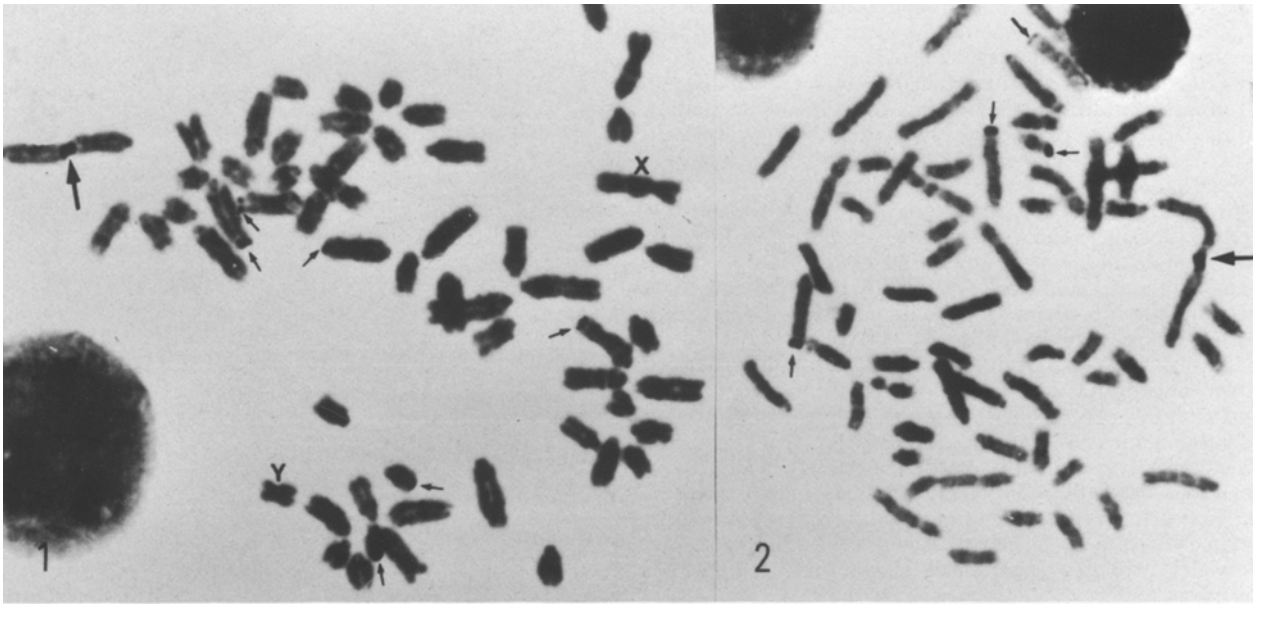

Fig. 1. Metaphase chromosomes of cattle (*Bos taurus*, 2n = 60) from Ag-G preparations. Small arrows indicate NOR sites on the telomeric regions of several chromosomes. Strong arrow indicates association between 2 NOR-bearing chromosomes. Fig. 2. Ag-G preparation from goat (*Capra hircus*, 2n = 60), showing chromosomes with Ag-NORs (small arrows). Association of 2 NOR chromosomes can be seen (strong arrow).

was dipped into this drop. The progress of staining must be followed under the microscope because NORs are stained immediately (20–40 sec). Time of developing can be delayed by using cold agents (+5 °C). The chromosomes remain a light yellow colour and the NORs stain black. After Ag-staining the preparations were rinsed in distilled water and dehydrated in 96% ethanol. For G-banding a pretreatment in phosphate buffer (pH 6.8) for 10 min was necessary. Thereafter the preparations were trypsinized in 0.025% trypsin (Difco) diluted in phosphate buffer at pH 6.8, for 5–7 min. Then the slides were rinsed in distilled water and stained in phosphate-buffered (pH 6.8) Giemsa (Merck) 1:15 for 8 min at room temperature. After rinsing in distilled water, the slides were dried and mounted in DPX.

The results of this Ag-G-staining can be seen in figures 1 and 2, which represent metaphases of cattle and goat. The chromosomes show a distinct banding pattern and the NORs at telomeric positions are clearly visible. As in humans[9], polymorphic behaviour of Nor bearing chromosomes could be observed, with variation in the number and amount of silver precipitates from individual to individual. This may be the expression of activity of rRNA which represents a Ag-NOR pattern characteristic for each individual and each species.

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An improved vacuum collector for fungal spores

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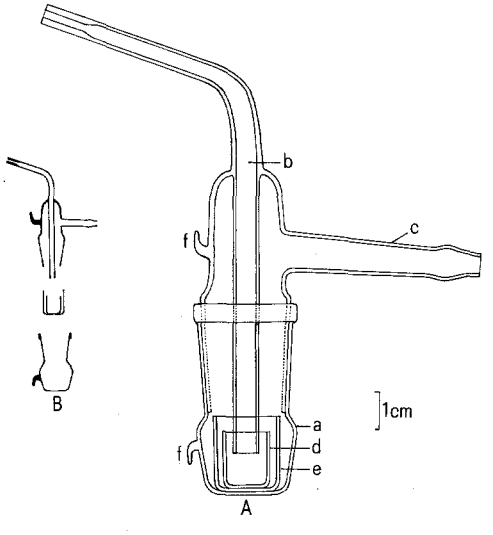
Summary. An all-glass improved vacuum collector for fungal spores is described and its merits mentioned. The description of the required components is also given. This vacuum collector combines the merits of smaller dimension and greater efficiency by accommodating Conway Diffusion cells in the spore receiving vessel.

The collection of dry fungal spores from infected plants or from solid culture surfaces is tedious and time-consuming; particularly when substantial quantities of material are required. In addition, the spores must be collected free of contaminants for the purpose of physiological study. For collection of dry fungal spores, small and large cyclone separators have been described[2,3]. Tervet et al.[4] described the use of a group of cyclone separators for collections of dry fungal spores from infected plants. A modified and enlarged version of the collector of Tervet et al.[4] has been described by Cherry and Peet[2] that permits rapid collection of fungal spores from cereal plants growing in pots, flats, or in the field. Woodbury et al.[3] have described a simple all-glass vacuum collector for collection of small quantities of fungal spores.

During the course of our studies on the physiology of fungal spore germination we have constantly felt the need for a simple all-glass vacuum spore collector that would be easy to operate, could be conveniently sterilized and could also provide spores free from contaminations. Consequently we report here an improved version of the collector described by Woodbury et al.[3]. Though originally deviced for collection of dry fungal spores from solid culture surfaces, this spore collector can be adapted for collection of pollen grains and spots from TLC plates.

The figure shows details of the construction. The list of materials shows the components and the number required. All materials are Jena Glass except the rubber band on the retainer used to keep the collector intact during operation. A water aspirator pump (not shown in the figure) is sufficient to provide the necessary vacuum.

Experience gained with the use of this spore collector has shown some distinct merits over the existing ones. This small glass collector can be conveniently sterilized. The Conway diffusion cells that can be accommodated inside the spore receiver permit collection of larger quantities of spores when required, simply by replacing them with other such cells or the micro-Conway diffusion cell if smaller quantities of spores need collection. Thus the need to have a larger spore-receiving vessel and thereby to increase the dimensions of the collector is eliminated. The spores while still inside the spore collector can be vacuum infiltrated



The spore collector, *A* assembled, and *B* dismantled, to show the components.