furanosyl residues[32]. It is believed that R.R. and Con-A do not specifically identify the same macromolecules[33].

In our experiments, changes observed in the thickness of the cell coat as visualized by Con-A permitted us to conclude that ovariectomy and cigarette smoke associated with ovariectomy modified glycoproteins on the surface of the thoracic aorta. We were unable to determine whether these changes at the molecular level were of a qualitative or quantitative nature. Also, we cannot be sure that cigarette smoke in itself did not affect to cell coat. These cell surface modifications are probably very important for explaining the increase in permeability during the initial stages of experimental arteriosclerosis since the integrity of the endothelial cell plasma membrane is directly dependent on the quantity and quality of structural glycoproteins in the cell coat[34].

Furthermore, these findings are important because they emphasize that hormonal factors can be related to arteriosclerosis and this may explain the higher rate of the disease in men.

1 Acknowledgment. This study was supported by grants from the Medical Research Council of Canada, the Canadian Heart Foundation and the J.C. Edwards Foundation.
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0014-4754/83/010072-03$1.50+0.20/0

Cyst formation in metanephric organ culture induced by cis-dichlorodiammineplatinum (II)

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Summary. A new experimental model of renal tubular cyst formation has been developed in metanephric organ culture. The addition of cis-dichlorodiammineplatinum (II), 50 μg/ml, to culture medium induces cystic changes during in vitro nephrogenesis. The model has particular utility in the study of basic mechanisms underlying renal tubular cystic changes, as well as the mechanisms by which nephrotoxins may mediate renal tubular injury.

The pathogenesis of human renal polycystic disease remains unknown despite numerous experimental studies in animal models[3,4]. We have recently described a mouse metanephric organ culture system for the study of normal renal development in which organotypic tubulogenesis and glomerular epithelial formation occur without capillary ingrowth or the presence of endothelial or mesangial elements[5]. Drawing on a recent report of renal cyst formation induced in adult rats by the i.v. administration of cis-dichlorodiammineplatinum (II) (CP)[6], we have utilized CP to induce cystic changes in the serum-free organ culture model. We have thus developed an experimental model in which renal polycystic changes occur during in vitro nephrogenesis without vascularization, glomerular filtration, or tubular urine formation.

Materials and methods. Our method of whole metanephric organ culture has been described in detail[5]. Pregnant Swiss-Webster albino mice are sacrificed by cervical dislocation at 13±0.4 days gestation. Under aseptic conditions fetal metanephric tissue is microdissected from embryos and transferred to a 0.8 μm Millipore filter sitting atop a Trowell double-welled organ culture assembly. Culture medium, which consists of equal volumes of Dulbecco's modified essential media and Ham's F-12 medium, supplemented with insulin (5 μg/ml), PGE-1 (25 ng/ml), T_3 (3.2 pg/ml), hydrocortisone (5 μg/ml), and transferrin (5 μg/

ml), is added, and the entire assembly incubated at 36±0.2 °C and 95% humidity in a mixed air −5% CO_2 environment. Culture medium is changed every 48 h. Following 24 h of normal incubation, CP (50 µg/ml) is added to tissue of the treatment group for 4 h. Culture medium alone is added to tissue of the control group. Following the 4-h period of drug treatment, all tissue is washed and returned to standard culture condition. Tissue from both drug therapy and control groups is then sampled daily for histological analysis and viability measurements as previously described[5].

Results. 48 h following CP treatment, mild tubular dilatation is noted amidst a background of normal in vitro nephrogenesis (fig. 1,a). Control tissue shows normal in vitro nephrogenesis as defined for the model system (fig. 1,b)[5]. 120 h post drug treatment, enlarged cystic tubules have formed among normally-differentiated elements (fig. 2,a). Control tissue at the same stage shows normal in vitro nephrogenesis with the formation of mature tubules and glomeruli composed totally of epithelial elements (fig. 2,b). Cellular viability of both treatment and control groups remained above 80% during the 6-day culture period.

Discussion. Current theories of the pathogenesis of renal tubular cystic changes focus on 2 major mechanisms: 1. obstruction to urine flow causing increased hydrostatic pressure and distention of normal structures[7,8]; and 2. toxic or metabolic injury causing damage to tubular supporting walls and distention of weakened structures[9]. Since previous experimental studies of cystic disease have utilized in vivo animal models, it has been difficult to experimentally isolate and study each of these potential pathogenetic mechanisms separately[3]. This study shows that renal cystic changes can be induced in developing renal tissue without vascularization, glomerular filtration, or urine formation. Since nephron obstruction and increased intratubular hydrostatic pressure cannot occur in the nonperfused metanephric organ culture system, the model isolates tubular supporting wall injury and resultant cystic changes for further experimental study.

CP is a potent chemotherapeutic agent whose clinical use is accompanied by a high incidence of acute and chronic renal tubular injury[10]. The precise mechanism of CP nephrotoxicity is unknown. Various theories advanced to explain its tubular toxicity include binding of sulfhydryl groups in proteins and nucleic acids, interfering with tubu-

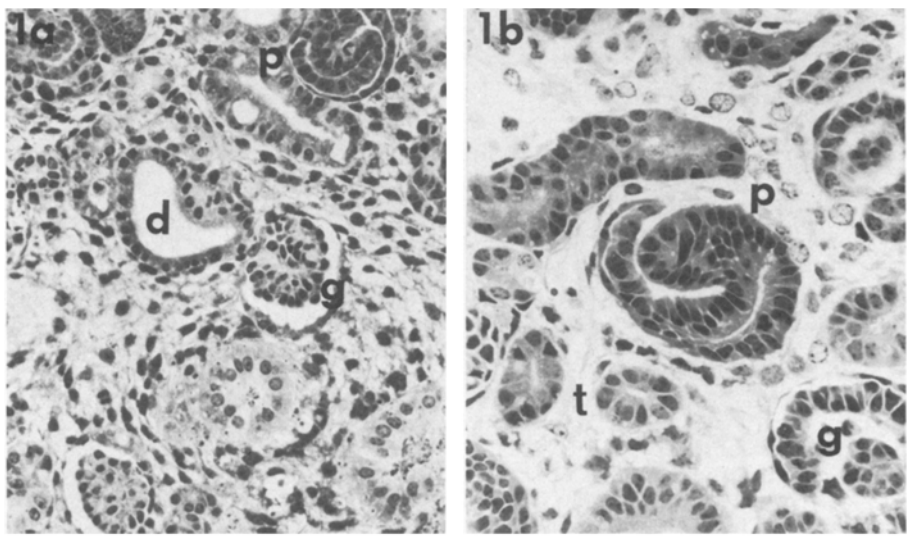

Figure 1. *a* CP-treated tissue 48 h post exposure shows mild tubular dilatation (d) amidst epithelial glomeruli (g) and pre-glomerular forms (p). (H&E×120). *b* Control in vitro tissue 48 h post exposure shows wellformed tubules (t), epithelial glomeruli (g), and pre-glomerular forms (p). (Trichrome × 150).

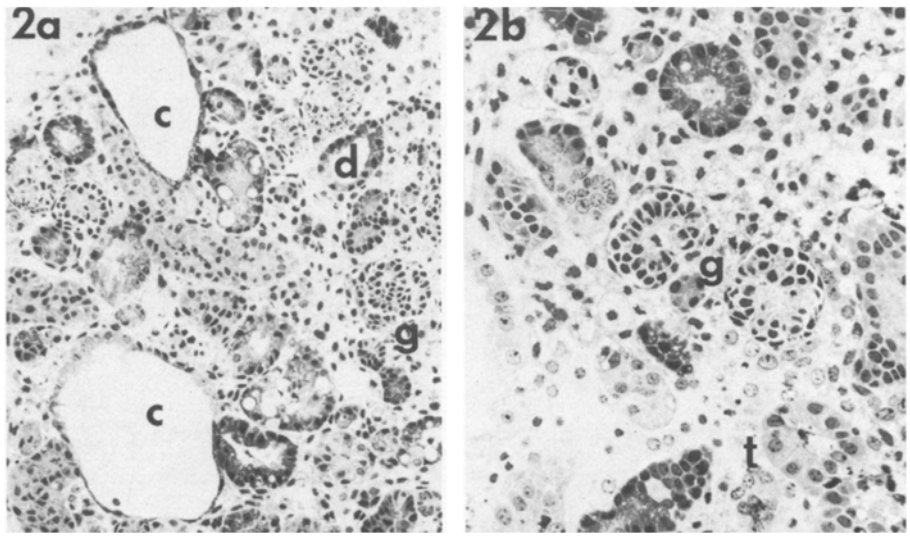

Figure 2. *a* CP-treated tissue 120 h post-exposure shows marked cystic tubular dilatation (c) amidst mildly dilated tubules (d) and normal epithelial glomeruli (g). (H&E×72). *b* In vitro controls at 120 h post-exposure show normal organ culture tubules (t) and glomeruli (g). (Trichrome × 150).

lar cell DNA synthesis, and inhibiting tubular ATP-ase and other transport enzymes[11]. In addition to its utility in the study of the pathogenesis of renal cystic disease, the organ culture model may have particular usefulness in the study of mechanisms by which tubular changes are induced by CP. The model isolates tubular toxicity from the influences of vascularization and permits a degree of experimental control which cannot be attained with in vivo models. Further, the use of completely defined serum-free medium in the organ culture model permits precise analysis of the biochemical changes which may mediate CP nephrotoxicity. Current studies are under way to define the biochemical changes which may precede or mediate CP-induced cystic changes in this model.

1 Acknowledgments. The authors thank Dr Jay Bernstein for his helpful suggestions. This work was supported by the National Kidney Fund of Western Pennsylvania and the Ronnie Miller Research Fund.
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0014-4754/83/010074-03$1.50+0.20/0

Colchicine induced interchanges in chillies (*Capsicum annuum* L.)

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Summary. Seeds of *Capsicum annuum* L. cultivar cerasiformies were treated with 0.4 and 0.2% aqueous colchicine solution for 24 and 72 h respectively. Tetraploids were not realized; instead, interchange heterozygosity was observed in several plants in 0.4% treatment. The interchanges varied from 1 to 3 per plant. It is presumed that colchicine has induced chromosome breaks.

Colchicine as a polyploidizing agent in various taxa has been well established. The fact that it also possesses mutagenic properties was first observed in *Sorghum*[2-7]. It was also noticed that this chemical has mutachromosomic properties[8], as first reported in chillies[9], in which a single interchange resulted with less than 50% pollen sterility but with good seed setting. Subsequently similar effects were reported in *Collinsia*[10], rye grass[11], and castorbeans[12]. The present study is a report of multiple chromosome interchanges induced by colchicine in *Capsicum annuum* L. cultivar cerasiformies.

Seeds of *C. annuum* were obtained from Government Agricultural Research Station Lam-farm, Guntur, Andhra Pradesh, India. To obtain polyploidy, 2 seed samples of 40 each were treated wtih 0.4 and 0.2% of aqueous colchicine (E. Merck, FRG) for 24 and 72 h respectively. Out of the 80 seeds thus treated, only 25 seeds germinated and grew to normal size (13 in 0.4% and 12 in 0.2%). Cytological screening of these plants revealed that polyploidy was not induced in any one of the surviving plants. Instead, interchange heterozygosity was noticed in 9 plants out of the 13 that were treated with 0.4% colchicine. Out of the 9 plants with interchanges, 3 interchanges each were recoreded in 2 plants, 2 interchanges in 2, while a single interchange was present in the remaining 5 plants.

Frequency of colchicine induced chromosomal interchanges at metaphase I in *Capsicum annuum* L.

Plants	Total No. of PMCs	Percentage of PMCs with interchanges						
		3 interchanges	2 interchanges	1 interchange	12 IIs	11 IIs+2 Is	10 IIs+4 Is	Pollen sterility (%)
1	120	50.00	20.00	10.00	5.00	3.33	11.66	83.20
2	110	59.09	16.36	14.54	1.81	-	8.18	88.50
3	90		60.00	13.33	15.55	-	11.11	82.00
4	90		66.67	12.22	4.44	11.11	5.55	83.80
5	100			75.00	6.00	8.00	11.00	80.10
6	90			64.44	20.00	7.78	7.78	76.90
7	100			73.00	15.00	4.00	8.00	78.00
8	110			63.64	18.18	10.00	8.18	76.80
9	71			100.00	-	-	-	84.70