12. Schneider, W., *J. Biol. Chem.*, 1945, v161, 293.
13. Schmidt, G., Thannhauser, S., *ibid.*, 1945, v161, 83.
14. Lineweaver, H., Burk, D., *J. Am. Chem. Soc.*, 1934, v56, 658.
15. Zamenhof, S., Reiner, B., De Giovanni, R., Rich, K., *J. Biol. Chem.*, 1956, v219, 165.

Received July 14, 1961. P.S.E.B.M., 1961, v108.

Thromboplastic and Fibrinolytic Activities of Large Human Vessels.* (26941)

S. COCCHERI AND TAGE ASTRUP

James F. Mitchell Foundation, Washington Clinic, Washington, D.C.,† Biological Inst., Carlsberg Foundation, Copenhagen, Denmark and Instituto di Patologia Speciale Medica, University of Bologna, Italy.‡

Intima and media of the human aorta are highly thromboplastic with little or no fibrinolytic activity, while adventitia is less thromboplastic and highly fibrinolytic(1). Hence, after tissue injury fibrin can be easily deposited on the intimal surface. Its removal, however, depends upon a fibrinolytically active system in blood. We have now investigated some large human vessels to elucidate further the physiological role of the hemostatic balance in the organism.

Experimental. Materials and methods were similar to those previously described (1). Thromboplastic activity was estimated on freshly drawn citrated rabbit plasma and compared with a suspension of human brain used for prothrombin estimations. Fibrinolytic activity was estimated after extraction with KSCN and expressed in units of a standard preparation of plasminogen activator prepared from pig heart. The fibrin plate method with 0.12% bovine fibrinogen clotted with bovine thrombin (Leo Pharmaceutical Prod., Copenhagen) was used. All estimations (thromboplastic and fibrinolytic) were made on serial dilutions of the samples.

Samples of the vessels were obtained from autopsies (21) at the Inst. of Forensic Medicine, Univ. of Copenhagen (with kind permission of Prof. H. Gormsen). Most of the samples came from subjects who had died from accidents. As in our previous report apparently normal or only slightly pathological specimens were compared. Complete separation of layers (under water) was obtained for arteries only. In veins media and adventitia could usually not be separated and the activities had to be estimated together. No effort was made to separate the endothelium from the intima proper, since nearly identical activities had been found in our previous study.

Results. The thromboplastic activity in the different layers of the vessels appears in Table I. In all vessels thromboplastic activity was low in adventitia. In the pulmonary vein and inferior vena cava it was also low in the intima. It reached medium values in the media and intima of the pulmonary artery. As before aortic intima was nearly always highly active. None of the curves indicated presence of antithromboplastic material. The activities of aortic and coronary intima in the same individuals were estimated in 5 subjects (Table II). The distribution follows identical patterns and the coronary intima has even higher thromboplastic activity than the aortic intima.

Estimations of plasminogen activator concentrations in 14 subjects are presented in Table III. Fibrinolytic activity was found in the aortic intima in only 4 cases, and in only one was there more than a trace of activity. It is perhaps significant that the sample with highest intimal activity (44.4

* This work was supported by the Josiah Macy Jr. Foundation and by a grant from U. S. Public Health Service, Nat. Heart Inst. The participation of S. C. was made possible by a Danish-Italian Exchange Fellowship.

† Present address of Dr. Astrup.

‡ Present address of Dr. Coccheri.

TABLE I. Thromboplastic Activities in Layers of Large Human Vessels (Expressed as Fractions of Activity of a Human Brain Preparation).

Age & sex	Cause of death	Aorta*			Pulm. artery			Inf. vena cava		Pulmonary vein	
		Int.	Med.	Adv.	Int.	Med.	Adv.	Int.	Med. + Adv.	Int.	Med. + Adv.
8 ♀	Cerebellar haemorrhage	1/21	—	<1/128	1/64	1/16	<1/128	1/100	<1/128	1/120	<1/128
35 ♂	Accident	2/3	—	—	1/6	—	—	1/14	—	1/14	—
35 ♂	CO poisoning	1/2	1/2	<1/128	1/16	1/8	<1/128	1/64	—	—	—
36 ♀	Uremia	1/2	1/4	<1/128	1/12	1/15	<1/128	<1/128	<1/128	<1/128	<1/128
38 ♂	Accident	1/4	—	—	1/32	1/16	1/64	—	—	1/50	<1/128
38 ♂	Pulm. emb.	1/6	1/3	<1/128	1/6	1/3	<1/128	1/32	<1/128	<1/128	<1/128
38 ♂	Uremia	1/6	1/12	1/32	—	—	—	—	—	—	—
41 ♀	Accident	1/16	1/28	<1/128	1/40	—	—	<1/128	<1/128	<1/128	<1/128
45 ♀	Spinal cord cancer	1/2	1/12	<1/128	1/10	1/14	<1/128	1/50	<1/128	1/40	<1/128
63 ♂	Coronary thrombosis	1/5	3/4	1/3	—	—	—	—	—	—	—
64 ♀	Accident	1/8	—	—	1/80	—	<1/128	<1/128	—	<1/128	—
70 ♀	Cerebral arteriosclerosis	1/10	1/20	<1/128	1/40	1/100	<1/128	—	—	—	—
73 ♂	Accident	1/4	1/8	<1/128	1/64	1/25	<1/128	1/80	<1/128	1/100	<1/128
	Avg	1/4	1/4	<1/128	1/16	1/10	<1/128	1/50	<1/128	1/60	<1/128

* Int. = intima; Med. = media; Adv. = adventitia.

units) was from a case of CO-poisoning since the only sample with measurable activity in our previous study (Ref. 1, Table III, 35 units) was also CO-poisoned. Aortic media showed trace activity in only one case. In 5 cases intima of the pulmonary artery had no fibrinolytic activity. The remaining 9 samples showed low activity. Media of the pulmonary artery was always active and was high in a single sample. Adventitia always had medium or high activity. All layers of the inferior vena cava and pulmonary vein were fibrinolytically active. Samples of aorta and coronary artery from the same individuals are compared in Table IV. The most interesting finding is that all 3 layers

TABLE II. Thromboplastic Activities of Intimal Layer of Coronary Arteries and Aortas of 5 Individuals. (Recorded as in Table I.)

Case No.	Aorta	Coronary artery
18	2/3	1/1
21	2/5	3/5
22	1/12	1/4
23	1/12	1/4
24	1/5	1/2
Avg	1/3	1/2

of the coronary artery have fibrinolytic activity, though intima is only slightly active.

Discussion. The potential ability of the arterial wall to form and dispose of fibrin deposits is of interest in study of the pathogenesis of arteriosclerosis, since fibrin formation on the inner arterial wall is the basis of the thrombogenic theory. Hence, several investigations on thromboplastic and fibrinolytic activity of arteries and veins have appeared recently.

Witte(2) observed high thromboplastic activities in the human arterial wall, while veins had lower activity. Kirk(3) confirmed the high thromboplastic activity of the human arterial intima. He found high activity in intima of vena cava, while intima from the pulmonary artery was lower. Stevenson *et al.* (4), using a different technic, found little thromboplastic activity in aortic intima, though it could substitute for platelets in the thromboplastin generation test. Todd(5), using a histological method, found fibrinolytic activity to be related to veins, while systemic arteries were inactive. In lung, fibrinolysis was produced by the pulmonary arteries and arterioles. In large arteries the vasa vasorum were active. Lieberman and Kellogg(6) confirmed the absence of plasminogen activator from arterial intima and its high concentration in adventitia.

The observation by these authors and the results of the present investigation show general agreement, though there are discrepancies. In contrast to Kirk(3) we found only

TABLE IV. Plasminogen Activator Concentration in Coronary Arteries and Aortas of 5 Individuals. (Recorded as in Table III.)

	Aorta			Coronary artery		
Case No.	Int.	Med.	Adv.	Int.	Med.	Adv.
15	0	0	456	13	48	810
20	10	3.8	432	18	27	918
21	6.2	2.7	280	19	36	583
23	0	0	385	0	5.3	172
24	0	0	296	11	29	324
Avg	3.2	1.3	370	12	29	561

TABLE III. Plasminogen Activator Concentration in Layers of Large Human Vessels (Expressed as Units/g Fresh Tissue).

			Aorta			Pulm. artery			Inf. vena cava			Pulm. vein	
Age & sex		Cause of death	Int.	Med.	Adv.	Int.	Med.	Adv.	Int.	Med.	Adv.	Int.	Med. + Adv.
4/12	♀	Accident	0	0	264	0	—	480	528	324		—	—
8/12	♀	Pneumonia	0	0	216	14	50	264	360	300	360	252	72
1	♂	Accident	0	0	552	26	122	480	240	264	1080	—	—
8	♀	Cerebellar haemorrhage	0	0	420	0	72	552	732	420	420	264	312
35	♂	CO poisoning	44	0	576	67	324	576	264	672		264	744
35	♀	Pneumonia	0	0	97	0	35	64	75	345		48	75
36	♀	Uremia	0	0	88	12.5	36	132	114	480		93	149
38	♂	"	0	0	168	3	17	138	210	132	432	96	60
45	♀	Spinal cord cancer	4	0	240	7.4	50	240	108	126	396	19	72
45	♂	Myocardial infarction	3.8	0	122	6.7	37	276	330	144	504	264	264
63	♂	Coronary thrombosis	0	0	194	0	72	252	218	—	336	84	273
64	♀	Accident	9.4	0	610	34	54	430	159	124	129	86	215
70	♀	Cerebral arteriosclerosis	0	0	324	0	37	204	—	—	—	—	—
73	♂	Accident	0	3.8	302	8.1	15	270	65	140	140	32	102
		Avg	4.4	.3	298	12.8	71	311	262	206	422	137	213

low thromboplastic activity in intima of vena cava. Though we observed fibrinolytic activity in most of the samples of pulmonary arterial intima (9 of 14) intimal activity in most samples of pulmonary vein was considerably higher and none was zero. The discrepancy between this finding and the observation by Todd(5) could possibly be due to the fact that he investigated small arteries and veins present in sections of lung tissue. We could confirm with our extraction method his histological finding of fibrinolytic activity in the intima of large systemic veins (vena cava inferior), which in some cases was even higher than in the corresponding sample of adventitia.

Summary. Thromboplastic and fibrinolytic activity was estimated in layers of large human vessels. Arterial intima has higher thromboplastic and lower plasminogen activator concentrations (usually zero) than the intima of large systemic veins (vena cava inf.). In all vessels adventitia has high fibrinolytic activity.

1. Astrup, T., Albrechtsen, O. K., Claassen, M., Rasmussen, J., *Circulation Research,* 1959, v7, 969.
2. Witte, S., Bressel, D., *Fol. haemat.,* 1958, v2, 236.
3. Kirk, J. E., *Fed. Proc.,* 1961, v20, 90, (Abst.).
4. Stevenson, T. D., Schrodt, G. R., Reed, L. M., *Am. J. Med. Sci.,* 1961, v241, 632.
5. Todd, A. S., *J. Path. Bact.,* 1959, v78, 281.
6. Lieberman, J., Kellogg, F., *Circulation Research,* 1961, v9, 515.

Received July 17, 1961. P.S.E.B.M., 1961, v108.

Effects of Selective Ultraviolet Irradiation of the Nuclei of Living Cells.* (26942)

P. O'B. MONTGOMERY, F. VAN ORDEN, L. L. HUNDLEY, C. L. CHAPMAN AND J. E. COOK
Dept. of Pathology, University of Texas Southwestern Medical School, Dallas

The authors have reported some of the manifestations of ultraviolet irradiation damage in living cells(1,2,3). The effects reported concerned the nature of the response of the whole cell to ultraviolet irradiation, and the effects of selective ultraviolet irradiation of the cytoplasm of living cells. The purpose of this report is to extend the previous observations by recording an additional type of ultraviolet irradiation of a selected area of the cell. This report deals with the nature of the damage produced in intact living cells by selective ultraviolet irradiation of the nuclei of these cells.

Methods. We have previously reported the technic of ultraviolet flying spot television microscopy and time-lapse cinematography (4,5,6). Recently the authors have recorded improvements in this technic which permit the use of double flying spot microscopy illumination(7). In the experiments reported here the principle of double flying spot illumination was employed to illuminate the nucleus of the cell with light of a peak wave length of 2600 Å and to illuminate the cytoplasm of the same cell with light of a peak wave length of 4500 Å. Control cells in the same field were illuminated solely with light of 4500 Å wave length. Although it was impossible to irradiate the nuclei of the cells without including a small amount of cytoplasm in the irradiated area, it is felt by the authors that this amount of irradiated cytoplasm was so small in comparison to that which was protected from the irradiation that its effects were negligible.

No presently available technic could be applied to determine the dose of irradiation given to the nucleus of the cell.

The ultraviolet emitting scanner tube and the visible light emitting scanner tube—which in this case acted as the source of il-

* Supported by Atomic Energy Comm. Contract, Damon Runyon Memorial Fund for Cancer Research, Southwestern Medical Fn. and Tobacco Research Industry Committee.