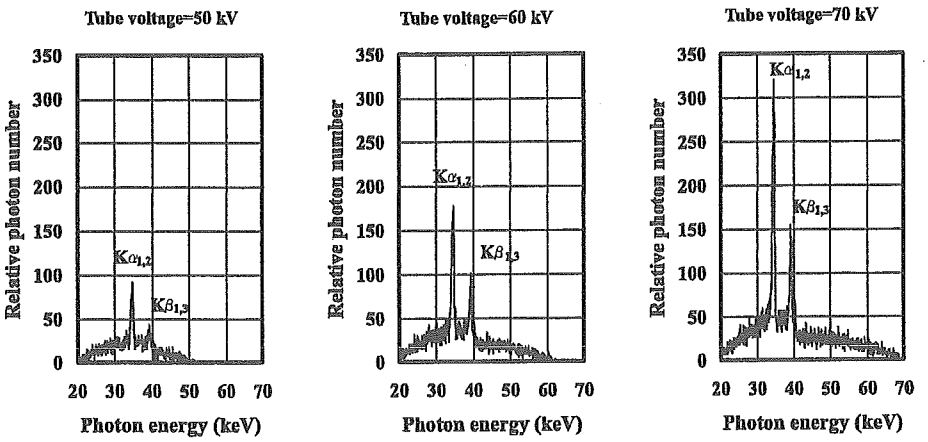

Fig. 4: X-ray spectra measured using a cadmium telluride detector with changes in the tube voltage.

system[21] with a sampling pitch of 87.5 μm. When the tube voltage was increased, spot dimensions increased slightly and had values of approximately 1×1 mm.

3.3 X-ray spectra

In order to measure x-ray spectra, we employed a cadmium telluride detector (XR-100T, Amptek Inc.) (Fig. 4). When the tube voltage was increased, the characteristic x-ray intensities of Kα and Kβ lines substantially increased, and both the maximum photon energy and the intensities of bremsstrahlung x-rays increased.

4. K-edge Angiography

Cerium is a rare earth element and has a high reactivity; however, the average photon energies of Kα and Kβ lines are 34.6 and 39.2 keV, respectively, and iodine contrast media with a K-absorption edge of

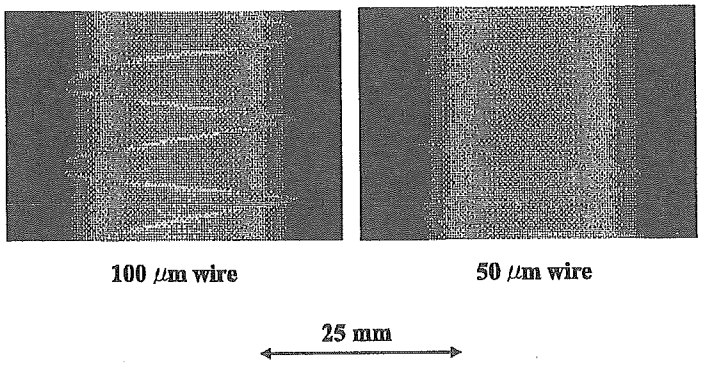

Fig. 5: Radiograms of tungsten wires coiled around PMMA rods.

33.2 keV absorb the lines easily. Therefore, blood vessels were observed with high contrasts.

The angiography was performed by the CR system[21] (Konica Regius 150) using the filter with a tube voltage of 60 kV, and the distance (between the x-ray source and the imaging plate) was 1.5 m. First, rough measurements of spatial resolution were made using wires. Figure 5 shows radiograms of tungsten wires coiled around rods made of polymethyl methacrylate. Although the image contrast decreased somewhat with decreases in the wire diameter, due to blurring of the image caused by the sampling pitch of 87.5 μm, a 50-μm-diameter wire could be observed.

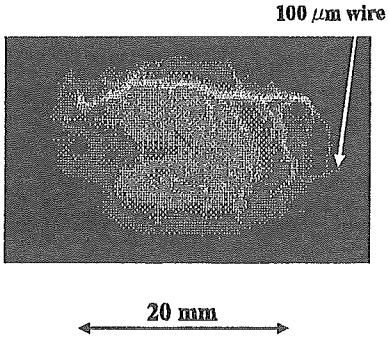

Fig. 6: Angiograms of an extracted rabbit heart using iodine microspheres.

An angiograms of a rabbit heart is shown in Fig. 6. This image was obtained using iodine microspheres of 15 μm in diameter. Fine blood vessels in the coronary arteries in the heart were visible. Figure 7 shows an angiogram of a larger dog heart using iodine spheres, and blood vessels of approximately 100 μm in diameter were visible.

5. Discussion

In summary, we employed an x-ray generator with a cerium-target tube and succeeded in producing cerium K-series characteristic x-rays, which can be absorbed easily by iodine-based contrast media. In the spectrum measurement, high-photon-energy bremsstrahlung x-rays beyond cerium K-edge (40.4 keV) were absorbed effectively.

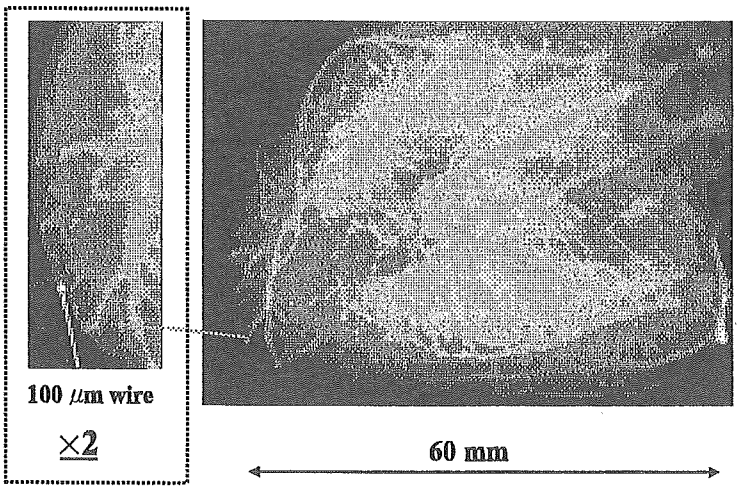

Fig. 7: Angiograms of an extracted dog heart using iodine microspheres.

In angiography, fine blood vessels were observed with high contrast with a spatial resolution of approximately 100 μm; the resolution was almost equal to the sampling pitch (87.5 mm) of the CR system. Therefore, the pith should be minimized, and magnification digital radiography including phase-contrast effect should be employed in order to improve the spatial resolution.

Although the cerium x-ray generator used in this research produces both the characteristic and the bremsstrahlung x-rays, bremsstrahlung intensity can be decreased effectively by considering the angle dependence without using the filter, since bremsstrahlung rays are not emitted in the opposite direction to that of electron trajectory. Subsequently, the generator produced maximum number of estimated characteristic photons was approximately 5×10^7 photons / (cm^3· s) at 1.0 m from the source, and the photon count rate can be increased easily by improving the target.

Acknowledgment

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Adrenomedullin: angiogenesis and gene therapy

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Nagaya, Noritoshi, Hidezo Mori, Shinsuke Murakami, Kenji Kangawa, and Soichiro Kitamura. Adrenomedullin: angiogenesis and gene therapy. *Am J Physiol Regul Integr Comp Physiol* 288: R1432–R1437, 2005; doi:10.1152/ajpregu.00662.2004.—Adrenomedullin (AM) is a potent, long-lasting vasodilator peptide that was originally isolated from human pheochromocytoma. AM signaling is of particular significance in endothelial cell biology since the peptide protects cells from apoptosis, promotes angiogenesis, and affects vascular tone and permeability. The angiogenic effect of AM is mediated by activation of Akt, mitogen-activated protein kinase/extracellular signal-regulated kinase 1/2, and focal adhesion kinase in endothelial cells. Both AM and its receptor, calcitonin receptor-like receptor, are upregulated through a hypoxia-inducible factor-1-dependent pathway under hypoxic conditions. Thus AM signaling plays an important role in the regulation of angiogenesis in hypoxic conditions. Recently, we have developed a nonviral vector, gelatin. Positively charged gelatin holds negatively charged plasmid DNA in its lattice structure. DNA-gelatin complexes can delay gene degradation, leading to efficient gene transfer. Administration of AM DNA-gelatin complexes induces potent angiogenic effects in a rabbit model of hindlimb ischemia. Thus gelatin-mediated AM gene transfer may be a new therapeutic strategy for the treatment of tissue ischemia. Endothelial progenitor cells (EPCs) play an important role in endothelial regeneration. Interestingly, EPCs phagocytose ionically linked DNA-gelatin complexes in coculture, which allows nonviral gene transfer into EPCs. AM gene transfer into EPCs inhibits cell apoptosis and induces proliferation and migration, suggesting that AM gene transfer strengthens the therapeutic potential of EPCs. Intravenous administration of AM gene-modified EPCs regenerate pulmonary endothelium, resulting in improvement of pulmonary hypertension. These results suggest that in vivo and in vitro transfer of AM gene using gelatin may be applicable for intractable cardiovascular disease.

regeneration; endothelium; ischemia; pulmonary hypertension

ADRENOMEDULLIN (AM) IS A POTENT, long-lasting vasodilator peptide that was originally isolated from human pheochromocytoma (36). The peptide consists of 52 amino acids with an intramolecular disulfide bond, sharing slight homology with calcitonin gene-related peptide and amylin. Immunoreactive AM is detected in plasma and a variety of tissues including, blood vessels, heart, and lungs (19). Particularly, AM shows a variety of effects on the vasculature that include vasodilatation (23), regulation of permeability (16), inhibition of endothelial apoptosis (31), and promotion of angiogenesis (1, 35, 60). In addition, AM has protective effects against vascular injury, including oxidative stress (33, 69, 84). It is becoming clear that either activation or disruption of AM signaling might contribute to many pathological conditions, including hypertension (22), congestive heart failure (55), pulmonary hypertension (29), neoplastic growth (39), and inflammatory disease (59). To date, the major biological acitivities of AM in vitro and in vivo are *1)* vasodilation, *2)* diuresis and natriuresis, *3)* positive inotropic effect, *4)* inhibition of endothelial cell apoptosis, *5)* induction of angiogenesis, *6)* inhibition of cardiomyocyte apoptosis, *7)* suppression of aldosterone production, *8)* anti-inflammatory activity, and *9)* antioxidant activity. We and others have demonstrated that intravenous administration of AM decreases systemic and pulmonary arterial pressure and induces diuresis and natriuresis (47, 52, 65), suggesting that AM is involved in the regulation of vascular tone and body fluid. Subsequent studies have demonstrated beneficial hemodynamic effects and direct cardioprotective effects of AM infusion in the treatment of congestive heart failure (57, 61–64).

Until recently, only vascular endothelial growth factor (VEGF) (80), fibroblast growth factor (68), platelet-derived growth factor (37), and angiopoietin (74) were known to have profound angiogenic effects. More recently, however, the angiogenic potential of AM has attracted investigators' attention (35, 41, 59, 81). A previous study has shown that vascular abnormalities are present in homozygous AM knockout mice (70), suggesting that AM is essential for vascular morphogenesis. AM activates the PI3K/Akt-dependent pathway in vascular endothelial cells (58), which is considered to regulate multiple critical steps in angiogenesis, including endothelial cell survival, proliferation, migration, and capillary-like structure formation (27). These findings raise the possibility that AM plays a role in modulating angiogenesis and neovascular-

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ization. This review focused on the angiogenic effects of AM and the therapeutic potential of AM gene transfer for the treatment of intractable cardiovascular disease.

ENDOGENOUS AM PRODUCTION IN ISCHEMIC CONDITIONS

Hypoxia (14, 53) and cytokine production (73) in ischemic heart disease or septic shock, as well as shear stress (7) in hypertension and heart failure induce AM secretion by vascular cells (Fig. 1). We have shown that plasma AM level is increased in patients with acute myocardial infarction (40, 49), peripheral arterial occlusive disease (75), and congestive heart failure (28, 55). Tissue levels of AM peptide and mRNA are also markedly increased in ischemic myocardium (18, 50) and failing heart (8, 56, 78, 82). These findings suggest that expression of AM is upregulated under tissue ischemia and inflammation, both of which are associated with neovascularization. An in vitro study has demonstrated that AM is upregulated through a hypoxia-inducible factor-1 (HIF-1)-dependent pathway under hypoxic conditions (14). Thus hypoxia/HIF-1 is one of the most potent regulators of AM production (Fig. 1). A recent study has demonstrated that heterozygous AM knockout mice [AM(+/−)] show significantly less blood flow recovery with less collateral capillary development than their wild-type mice (20). Administration of AM promotes blood flow recovery and capillary formation in AM(+/−) mice. These findings suggest that endogenous AM may play an important role in the regulation of angiogenesis under ischemic conditions. Considering the angiogenic potency of AM, increased endogenous AM represents a compensatory mechanism as an angiogenic factor promoting neovascularization under hypoxic conditions.

ANGIOGENIC EFFECTS OF AM AND ITS SIGNALING PATHWAY

AM signaling is of particular significance in endothelial cell biology since the peptide protects cells from apoptosis (31), promotes angiogenesis (35, 60), and affects vascular tone (23). Angiogenesis is a multistep process that involves migration and proliferation of endothelial cells, functional maturation of the newly assembled vessels, and remodeling of the extracellular matrix (26). Akt, mitogen-activated protein kinase (MAPK)/extracellular signal-regulated kinase 1/2 (ERK1/2), and focal adhesion kinase (p125FAK) play an important role in angiogenesis in endothelial cells. Kim et al. (35) demonstrated that AM activated Akt, MAPK/ERK1/2, and p125FAK in human umbilical vein endothelial cells (HUVECs), and produced increases in their DNA synthesis and migration. AM induced tube formation in HUVECs, and its effect was inhibited by pretreatment with a phosphatidylinositol 3′-kinase (PI3K) inhibitor or mitogen-activated protein kinase/extracellular signal-regulated kinase kinase (MEK)1/2 inhibitor. These findings suggest that AM exerts angiogenic activities through activation of Akt, MAPK, and p125FAK in endothelial cells (Fig. 1). In vivo, overexpression of AM augments collateral flow in ischemic tissues partly through activation of endothelial nitric oxide synthase (eNOS) (1). Earlier studies have shown that the vasodilatory effects of AM are mediated by cAMP/protein kinase in smooth muscle cells (SMCs) (23) and by the eNOS/NO pathway in endothelial cells (17). Thus AM-induced angiogenesis and vasodilation may synergistically improve blood perfusion in ischemic tissues.

Recently, a seven-transmembrane G-protein-coupled receptor, calcitonin receptor-like receptor (CRLR), and receptor activity modifying proteins (RAMPs) have been recognized as integral components of the AM signaling system (38, 43). CRLR has demonstrated the expression of the transcript predominantly in microvascular endothelial cells. This finding supports the view that CRLR is potentially a major mediator of the effects of AM on the vasculature. The effect of AM on CRLR is modified by RAMP2 and RAMP3. The angiogenic effect of AM is mediated by CRLR/RAMP2 and CRLR/RAMP3 receptors (Fig. 1). VEGF and AM act synergistically to induce angiogenic-related effects on endothelial cells in vitro (11). However, blocking antibodies to VEGF cannot significantly inhibit AM-induced capillary tube formation by

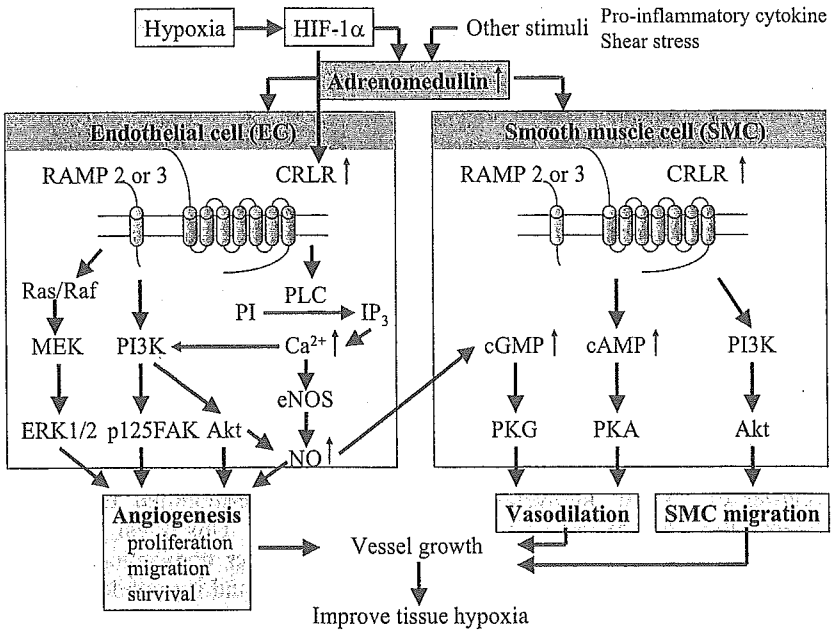

Fig. 1. Signaling pathway of adrenomedullin (AM) in vascular endothelial cells and smooth muscle cells. Both AM and calcitonin-receptor-like receptor (CRLR) are upregulated through a, hypoxia-inducible factor-1 (HIF-1)-dependent pathway under hypoxic conditions. AM binds to CRLR modified by receptor-activity-modifying protein 2 (RAMP2) and RAMP3. AM induces angiogenesis through activation of Akt, MAPK, and p125FAK in endothelial cells. AM also induces SMC migration and vasodilation. These activities synergistically improve tissue ischemia. MEK, mitogen-activated protein kinase/extracellular signal-regulated kinase kinase; ERK, extracellular signal-regulated kinase; PI3K, phosphatidylinositol 3-kinase; p125FAK, focal adhesion kinase; PLC, phospholipase C; PI, phosphatidylinositol; IP_3, inositol triphosphate; eNOS, endothelial nitric oxide synthase; NO, nitric oxide; cGMP, guanosine 3′,5′-cyclic monophosphate; PKG, protein kinase G; PKA, protein kinase A.

HUVECs, indicating that AM does not function indirectly through upregulation of VEGF. Interestingly, AM and CRLR are both upregulated under hypoxic conditions in microvascular endothelial cells, although expression of RAMPs is not activated by hypoxia in microvascular cells (54). The activity of the CRLR promoter under hypoxic conditions is regulated at least in part through hypoxia-responsive regulatory element binding transcription factor HIF-1. Thus the simultaneous transcriptional upregulation of CRLR and its ligand AM in endothelial cells might play a significant role in the vascular responses to hypoxia and ischemia by creating a potent survival loop.

SMCs are essential for the generation of functional and mature blood vessels (26). We demonstrated in vivo that intramuscular administration of AM increased the number of αSMA-positive cells involved in the formation of vascular structures (25). In vitro, AM enhanced SMC migration, which was inhibited by wortmannin, a PI3K inhibitor. Recent studies using homozygous AM knockout mice have suggested that AM is essential for vascular morphogenesis (6, 21, 70). Taking these findings together, it is possible that AM contributes to vessel maturation through enhancement of SMC migration via a PI3K/Akt-dependent pathway (Fig. 1). This feature of AM-induced angiogenesis is different from VEGF-induced angiogenesis, which is not associated with vessel muturation.

In tumor cells, inflammation and hypoxia increase AM expression, and the elevated expression of AM is associated with tumor neovascularization in xenografted endometrial tumors and renal cell carcinoma (12, 86). AM also acts as a tumor cell survival factor underlying human carcinogenesis. Thus hypoxia-induced AM plays a part in tumor angiogenesis in conjunction with VEGF, and facilitates tumor growth under hypoxic conditions. As angiogenesis is an essential process in tumor-host interactions for tumor growth, maintenance, and metastasis, finding ways to regulate the action of AM may provide a new avenue for developing anticancer therapy (16).

THERAPEUTIC ANGIOGENESIS

A variety of studies have demonstrated that AM gene delivery serves as therapeutic tool to protect the cardiovascular system, including the heart (9, 32, 85), kidney (83), and vasculature (2, 84). In this section, we describe the angiogenic potential of AM gene transfer using novel gene delivery systems.

Nonviral gene transfer. Peripheral vascular disease is a crucial health issue affecting an estimated 27 million people (5). Despite recent advances in medical interventions, the symptoms of some patients with critical limb ischemia fail to be controlled. Although gene therapy has been shown to be an effective approach for angiogenesis (10, 24, 72), it is still unsatisfactory because of the biohazard of viral vectors, low transfection efficiency, and premature tissue-targeting. Therefore, highly efficient and safe gene transfer is desirable. Recently, we developed a novel nonviral vector, gelatin hydrogel, which allows highly efficient and long-lasting gene transfer (13, 30, 81). Gelatin has been widely used as a carrier of protein because of its capacity to delay protein degradation (76, 77). Plasmid DNA is known to be negatively charged. Thus we used gelatin as a vector for gene therapy. Biodegradable gelatin was prepared from pig skin. The gelatin was characterized by a spheroid shape with a diameter of ~30 μm, water content of 95% and an isoelectric point of 9 after swelling in water (76, 77). After 2-h incubation, positively charged gelatin held negatively charged plasmid DNA in its positively charged lattice structure. DNA particles are released from the gelatin through its degradation. As a result, DNA-gelatin complexes can delay gene degradation, leading to efficient gene transfer (13, 30, 44, 81).

We examined whether nonviral vector gelatin-mediated AM gene transfer induces therapeutic angiogenesis in a rabbit model of hindlimb ischemia (81). Seven days after intramuscular injection of AM DNA-gelatin complexes, there was intense AM immunoreactivity surrounding the gelatin in the skeletal muscles. AM production in the AM-gelatin group was enhanced compared with that in the naked AM DNA group, which received plasmid AM DNA alone. Unlike AM production in the naked AM group, AM overexpression in the AM-gelatin group lasted for longer than 2 wk. Importantly, AM DNA-gelatin complexes induced more potent angiogenic effects in a rabbit model of hindlimb ischemia than naked AM DNA, as evidenced by significant increases in histological capillary density, calf blood pressure ratio, and laser Doppler flow. These results suggest that the use of biodegradable gelatin as a nonviral vector augments AM expression and enhances AM-induced angiogenic effects. AM DNA-gelatin complexes were distributed mainly in connective tissues. It is interesting to speculate that the delay of gene degradation by gelatin may have been responsible for the highly efficient gene transfer. Thus gelatin-mediated AM gene transfer may be a new therapeutic strategy for the treatment of severe peripheral vascular disease.

Cell-based gene transfer. Recently, transplantation of stem cells or progenitor cells has been shown to regenerate a variety of tissues. Endothelial progenitor cells (EPCs) have been discovered in adult peripheral blood (4, 79). EPCs are mobilized from bone marrow into the peripheral blood in response to tissue ischemia or traumatic injury, migrate to sites of injured endothelium, and differentiate into mature endothelial cells in situ (15, 34). Transplantation of EPC induces therapeutic angiogenesis in the ischemic heart or limb (34, 42, 71). However, some patients are refractory to conventional cell therapy because of insufficient cell number, poor survival, or impaired differentiation. Thus a novel therapeutic strategy to enhance the angiogenic properties of EPCs is desirable. Considering the variety of protective effects of AM on vascular endothelial cells, we hypothesized that AM gene transfer into EPCs would strengthen the therapeutic potential of EPCs. Genetically modified EPCs may serve not only as a tissue-engineering tool to reconstruct the vasculature but also as a vehicle for gene delivery to injured endothelium.

Here, we present a new concept for cell-based gene delivery into the vasculature, consisting of three processes (44). First, positively charged gelatin is readily complexed with negatively charged plasmid DNA. Second, EPCs phagocytose ionically linked plasmid DNA-gelatin complexes in coculture, which allows nonviral gene transfer into EPCs with high efficiency. Third, intravenously administered gene-modified EPCs are incorporated into injured vascular beds. This novel gene delivery system has great advantages over conventional gene therapy; it is nonviral and noninvasive, and it provides highly efficient gene targeting into the vasculature. These benefits

may be achieved mainly by the capability of EPCs to phagocytose DNA-gelatin complexes and to migrate to sites of injured endothelium. Genetically modified EPCs markedly secreted AM into the culture medium, and AM overproduction lasted for more than 2 wk. The proliferative activity of AM DNA-transduced EPCs exceeded that of nontransduced EPCs. Furthermore, AM gene transfer inhibited apoptosis of EPCs in vivo and in vitro. Thus ex vivo AM gene transfer strengthened the therapeutic potential of EPCs.

Primary pulmonary hypertension (PPH) is a rare, but life-threatening disease characterized by progressive pulmonary hypertension, ultimately producing right ventricular failure and death (67). Median survival in patients with PPH is considered to be 2.8 years from the time of diagnosis. Thus novel and effective therapy is needed for the treatment of pulmonary hypertension. Because endothelial dysfunction may play a role in the pathogenesis of pulmonary hypertension such as PPH (3), pulmonary endothelial cells may be a therapeutic target for the treatment of pulmonary hypertension. We have demonstrated that administration of AM peptide decreases pulmonary vascular resistance in patients with PPH (45, 46, 48, 51). Thus we investigated the effects of AM gene-modified EPCs on pulmonary hypertension in rats (44). AM gene-transduced EPCs were similarly incorporated into the pulmonary vasculature. Immunohistochemical analyses demonstrated that the transplanted EPCs were of endothelial lineage and formed vascular structures. Intravenous administration of AM-expressing EPCs significantly decreased pulmonary vascular resistance compared with EPCs alone (−39%). Kaplan-Meier survival curves demonstrated that rats with pulmonary hypertension transplanted with AM-expressing EPCs had a significantly higher survival rate than those given culture medium or EPCs alone. These findings suggest that AM gene-modified EPCs using gelatin may serve not only as a tissue-engineering tool to reconstruct the pulmonary vasculature, but also as a vehicle for gene delivery to injured pulmonary endothelium. This hybrid cell-gene therapy may be applicable for intractable cardiovascular disease, including ischemic heart disease. Thus genetic manipulation of stem cells opens new avenues for regenerative medicine.

GRANTS

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シンクロトロンにかわる医用単色X線装置の開発と応用

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Development of monochromatic x-ray generators instead of a synchrotron and applications

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1. はじめに

X線レーザーの研究は世界各国で行われており，レーザー発振のためのさまざまな方法が考案されている[1-4]。レーザーは誘導放出による光の増幅を意味するが，誘導放出によってはフォトンエネルギーを高めることは難しい。このことから誘導放出とは異なるが，自由電子レーザー[5]による方法が考案され，研究されている。人体をも撮影できるハードX線レーザーの発振はノーベル賞に値するといわれているが，まだそれらの発生は報告されていない。したがって，もし仮にハードX線レーザーが定常的に発生できれば，医療における診断や治療に大きく貢献すると思われる。

シンクロトロンとモノクロコリメーターを用いて発生する単色平行X線はヨウ素のKエッジ造影

[6-8] や位相コントラスト撮影 [9-11] 等に応用され，X線撮影上革命的な成果をあげている。特に，血管造影にはK吸収端が33.2 keVのヨウ素系造影剤が利用されるので，吸収端よりもわずかに高いフォトンエネルギーのX線は造影剤に効率良く吸収される。したがって，35 keV程度の単色平行X線は微小血管造影には非常に有用であることから，造影室はシンクロトロン施設内に設置されている。

マイクロフォーカスX線管を用いた輪郭強調X線位相コントラスト撮影は Wilkins[12] により考案された。最近，100 μm程度の小焦点モリブデン管とCRを用いた高精細マンモグラフィーシステム [13,14] がコニカミノルタから発売され，普及しつつある。X線撮像においてもデジタル化は進んでいるが，イメージングプレートを用いたコンピューターラジオグラフィー（CR）[15] やフラットパネルディテクター（FPD）はX線フィルムと比較して空間分解能が劣るので，拡大撮影による分解能の向上は必須である。さらに拡大により被写体からの散乱線の影響が低減され，位相コントラストの効果が加わる。このことから筆者等はデジタル拡大撮影の微小血管造影への応用を試み，良好な成果を得ている。

筆者等は単色X線撮影を行うため，エネルギー選択式のFPDの開発も行っているが，本稿では，近年開発した，種々のX線装置の特性やCRにより撮影した画像について簡単に説明する。

2. 低フォトンエネルギープラズマX線装置

低フォトンエネルギーの弱電離プラズマX線装置 [16-20] はハードX線レーザーの基礎研究のために開発され，銅やニッケル等のK系列特性X線を出力させるのに有用である。Fig. 1のように200 nFのコンデンサーを50 kV程度に充電し，蓄積された電荷をX線管の陰極にトリガ電圧を印加することにより放電する。この装置では高エネルギー放電により弱電離プラズマを成長させ，これを線状に形成することにより，制動X線が吸収され，蛍光X線（特性X線）に変換される。吸収係数が不連続なことから特性線はプラズマを容易に透過するので，単色化フィルターを挿入しなくとも高線量率の準単色X線が発生する。加えて，Kエッジフィルターにより$K\beta$線を吸収すれば，$K\alpha$線が得られる。X線管には長い棒状ターゲットが取付けてあり，1 mPa程度に連続排気される。

管電圧と電流は減衰振動となり，それらの最大値は充電電圧を高めることにより増加した。実験結果より，管電圧の最大値は充電電圧にほぼ匹敵し，最大管電流値は約15 kAであった。また熱蛍光線量計で測定した最大X線強度は線源から1.0 mの位置で1パルス当たり1.5 mGy程度であった。

X線スペクトルの測定には，厚さ0.5 mmのフッ化リチウム湾曲単結晶付の透過式分光器を用いた（Fig. 2）。実験ではクリーンなK系列特性X線が観測され，充電電圧の増加によりX線強度は著しく増加した。驚くことに，充電電圧が50 kVでは，高調波が観測された。

X線撮影には厚さ10 μmのニッケルフィルターを用い，撮影距離と充電電圧はそれぞれ1.2 mと50 kVであった。写真はプラスチックの試験管からこぼれ落ちるプラスチック弾である（Fig. 3）。この装置のX線照射時間は約1 μsであるため，完全静止画像が得られた。

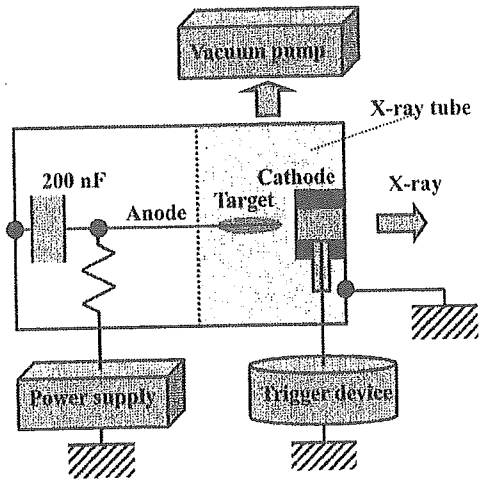

Fig. 1. Block diagram of the low photon energy plasma flash x-ray generator.

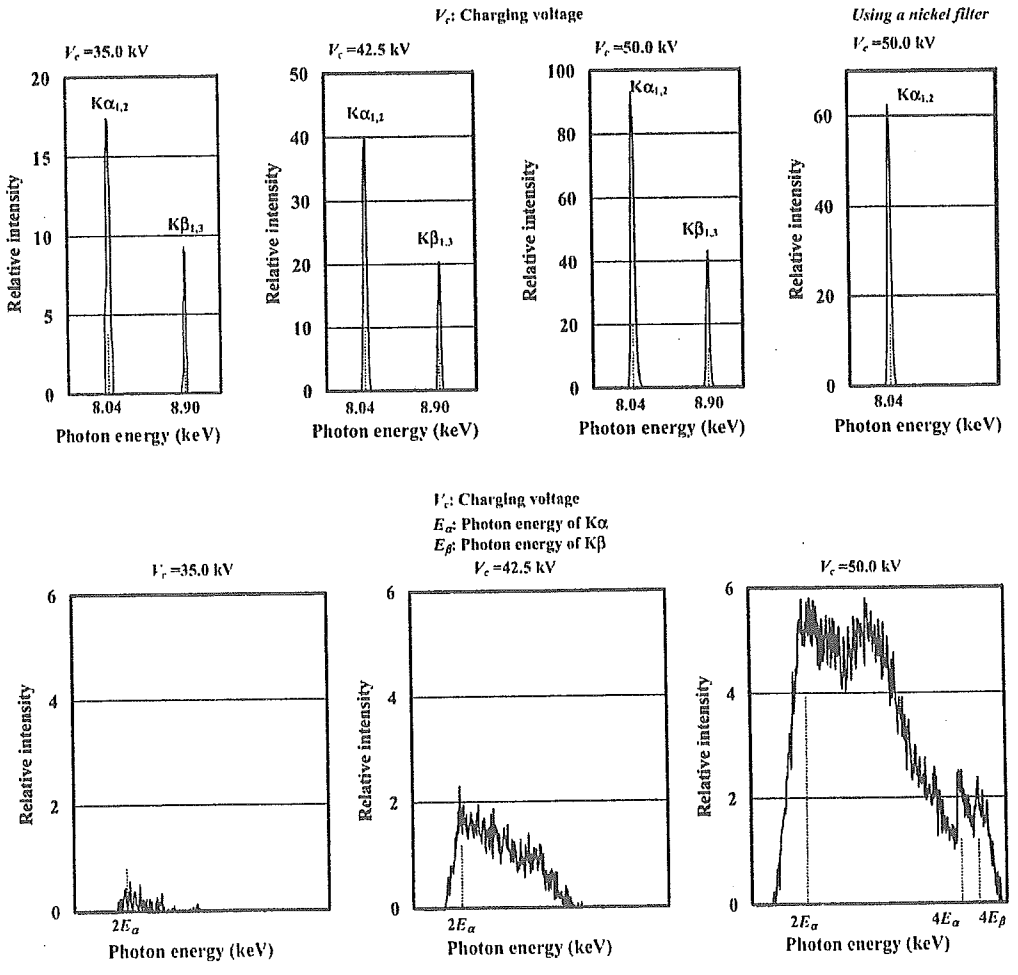

Fig. 2. X-ray spectra from weakly ionized linear plasma at the indicated conditions.

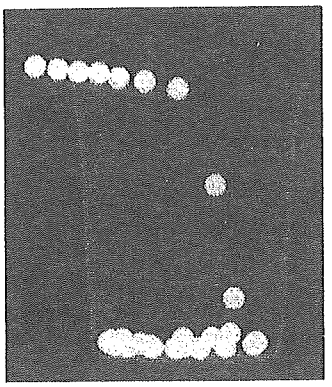

Fig. 3. Radiogram of plastic bullets falling into polypropylene beaker from a plastic test tube.

3. 制動X線の角度分布を用いた単色フラッシュX線装置

この単色フラッシュX線装置[21,22]は高フォトンエネルギーのクリーンな特性X線を発生させるために試作した。制動線は電子軌道と反対方向には出力し難い性質を利用した場合には，比較的容易に準単色あるいは単色のX線を得ることができる。この装置は高電圧パルス発生装置，ターボ分子ポンプ，X線管などからなる。パルス発生装置では2段マルクス回路を採用し，充電電圧の約−2倍の高電圧パルスが出力する。X線管にはグラファイト製の円盤状陰極と棒状のモリブデンターゲットが付いており，陰極表面からの電子ビームがターゲット先端に衝突し，X線は陰極とマイラーX線窓を透過して出力する（Fig. 4)。管体はアクリル製で，1 mPa 程度に連続排気される。スペクトルは前述の結晶分光器を用いて測定した。厚さ 20 μm のジルコニウムフィルターを用いた場合には Kβ線が吸収されるので，クリーンな単色の Kα線を得ることができた（Fig. 5)。フラッシュX線装置の管電圧と電流は減衰振動となり，それぞれの最大値は充電電圧が−70 kV の条件下で 120 kV と 1.0 kA であった。また熱蛍光線量計で測定した最大X線強度は線源から 0.5 m の位置で 1 パルス当たり 70 μGy であった。

Fig. 6 はガラス製試験管から流れ出る水で，撮影距離と充電電圧はそれぞれ 0.5 m と−70 kV であった。X線パルスの幅は約 70 ns であるため，水の完全静止画像を撮影できた。

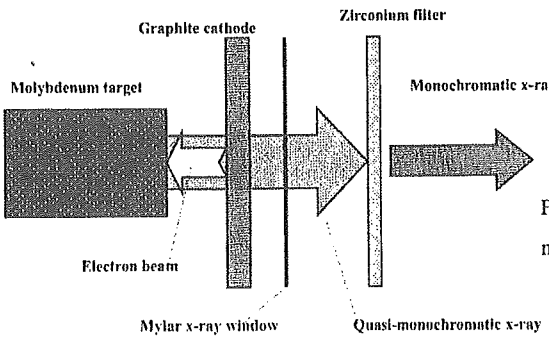

Fig. 4. K-photon irradiation using a monochromatic flash x-ray tube.

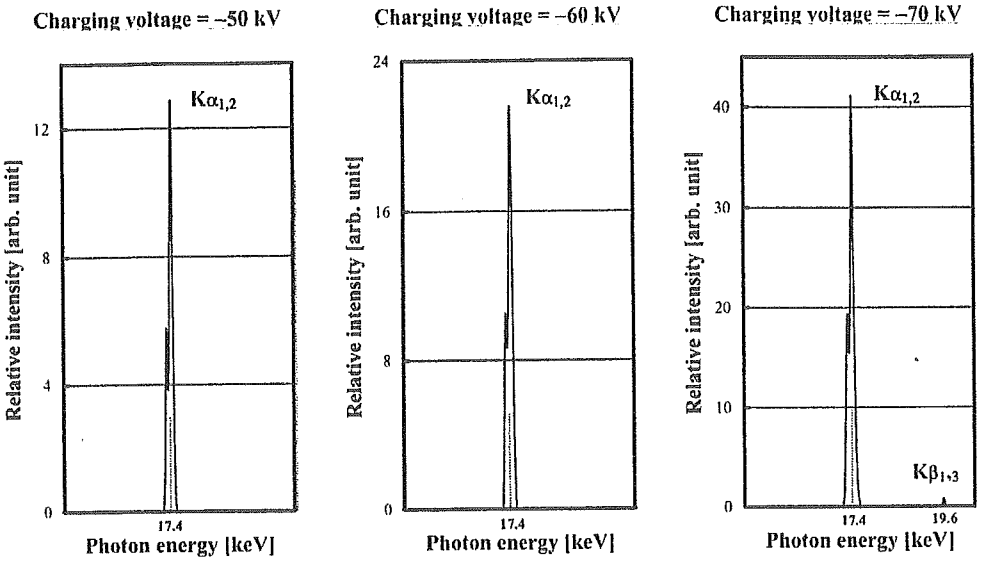

Fig. 5. X-ray spectra from a molybdenum target.

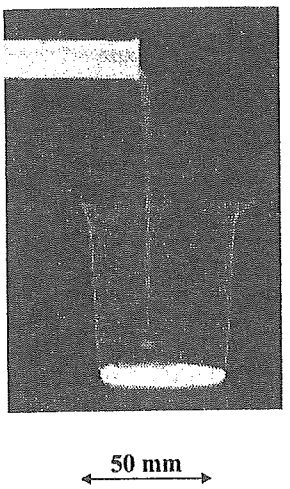

Fig. 6. Radiogram of water falling into polypropylene beaker from a glass test tube.

4. 高フォトンエネルギープラズマX線装置

高フォトンエネルギーのクリーンなK系列特性X線とそれらの高調波を発生させるため，高フォトンエネルギーのプラズマX線装置[23]を製作した。このフラッシュX線装置は高電圧充電器，コンデンサーユニット，ギャップスイッチ，ターボ分子ポンプ，そしてフラッシュX線管等よりなる。150 nFのコンデンサーを80 kV程度に充電し，蓄積された電荷をギャップスイッチを閉じることによりX線管内に放電し，フラッシュX線を得る。管はターボ分子ポンプにより1 mPa程度に連続排気される。

Fig. 7はK系列特性X線発生の原理図で，直径3.0 mmの棒状タンタルターゲットと内径4.5 mmのリング状グラファイト陰極が取り付けてある。陰極からの電子ビームはターゲット先端にほぼ垂直

に衝突するので，プラズマが形成され，図のようにK系列特性（準単色）X線が発生する。

ほぼ製品に近いプロトタイプゆえ，管電圧と電流の最大値を測定することは難しいが，充電電圧が80 kVの場合の最大値はそれぞれ160 kVと40 kA程度である。2極管を用いているので，X線照射時間は短く，約100 ns程度であった。次にフラッシュX線装置の線量率は極めて高いことから，半導体検出器を用いてスペクトルを測定することはできない。モリブデンターゲットから出力するX線スペクトルも結晶分光器を用いて測定したが，特性X線強度は充電電圧を高めることにより著しく増加した。しかし，20 μm厚のジルコニウムフィルターを用いてKβ線を吸収することは難しく，高調波も発生しなかった。一方，タンタルターゲットでも，ほぼクリーンなK系列特性X線を得ることができた（Fig. 8）。また熱蛍光線量計で測定した最大X線強度は線源から1.0 mの位置で1パルス当たり約300 μGyであった。

Fig. 9はガドリニウムの質量吸収係数とタンタルKα線の平均フォトンエネルギーの関係を示している。ガドリニウム造影剤はMRAで使用されるが，図のようにタンタルKα線はガドリニウムに効率よく吸収される。実験結果から，重量百分率で15 %程度の造影剤を用いれば，十分に高コントラストで撮影できる。酸化ガドリニウムを用いて造影したウサギ頭部をFig. 10に示す。図のように100 μm程度の微小血管が観察できた。

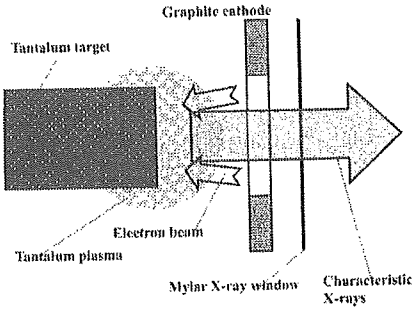

Fig. 7. K-photon irradiation from weakly ionized tantalum plasma.

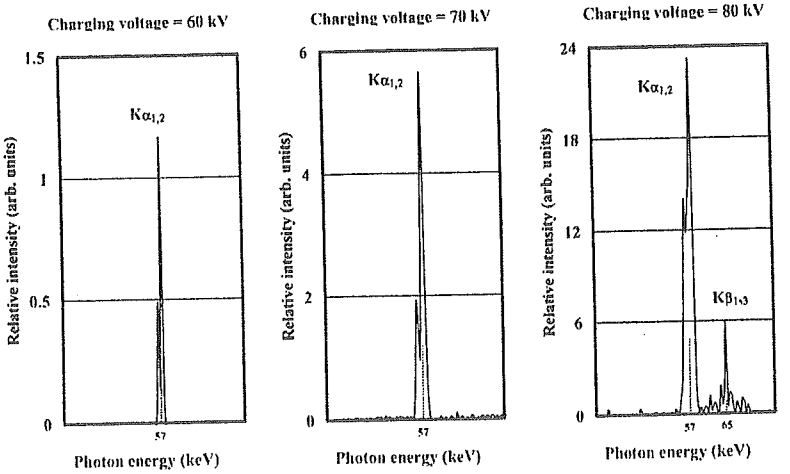

Fig. 8. X-ray spectra from a tantalum target.

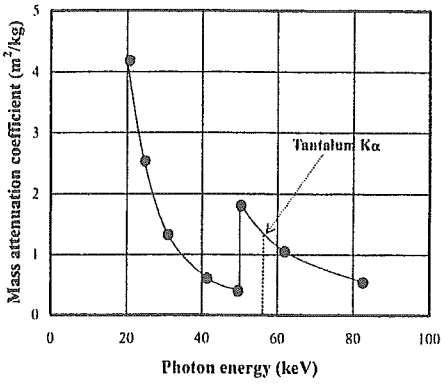

Fig. 9. Mass attenuation coefficient of gadolinium and the average photon energy of tantalum

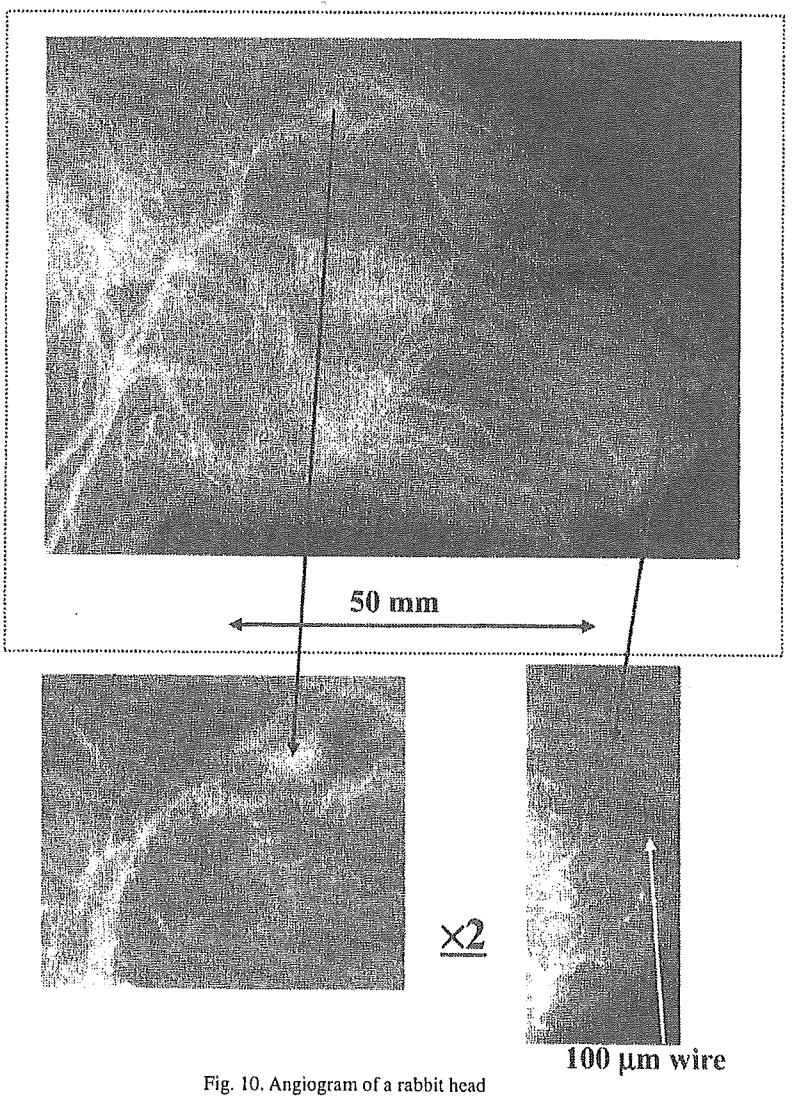

Fig. 10. Angiogram of a rabbit head

5. 制動X線の角度分布を用いた単色X線装置

定常的でクリーンなK系列特性X線を発生させるために，熱陰極（フィラメント）付きのX線管と制動X線の角度分布を用いた単色X線装置を製作した。この装置は高電圧電源，ターボ分子ポンプ，開放型のX線管等よりなる。管はターボ分子ポンプにより 0.5 mPa 程度に連続排気される。X線管の陽極には正の高電圧が印加され，陰極は接地されている。Fig. 11 はX線管の構造図で，フィラメントからの電子流は収束電極でターゲット先端に集められる。この実験では棒状ターゲットを使用したが，板状でも発生できる。管にはモリブデンターゲットを取付けたので，単色化のためのKエッジフィルターは 20 μm 厚のジルコニウムである。管電圧が 30 kV，管電流が 0.10 mA の条件におけるX線強度は，線源から 1.0 m の位置で 12.1 μGy/s で，クリーンな Kα線が観測できた (Fig. 12)。Fig. 13 はウサギの大腿で，直径 15 μm のヨウ素マイクロスフェアーを使って造影した。マイクロスフェアーは生体の造影ファントムを製作するのに有用である。フラッシュX線管と比較して，このX線管では高フォトンエネルギーのクリーンなK系列特性X線を得ることは難しい。

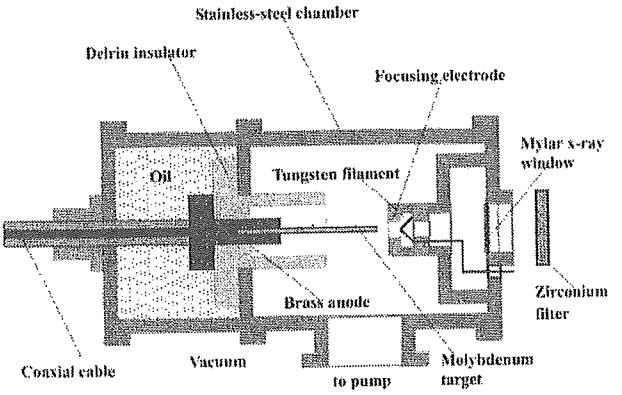

Fig. 11. Schematic drawing of the monochromatic x-ray tube.

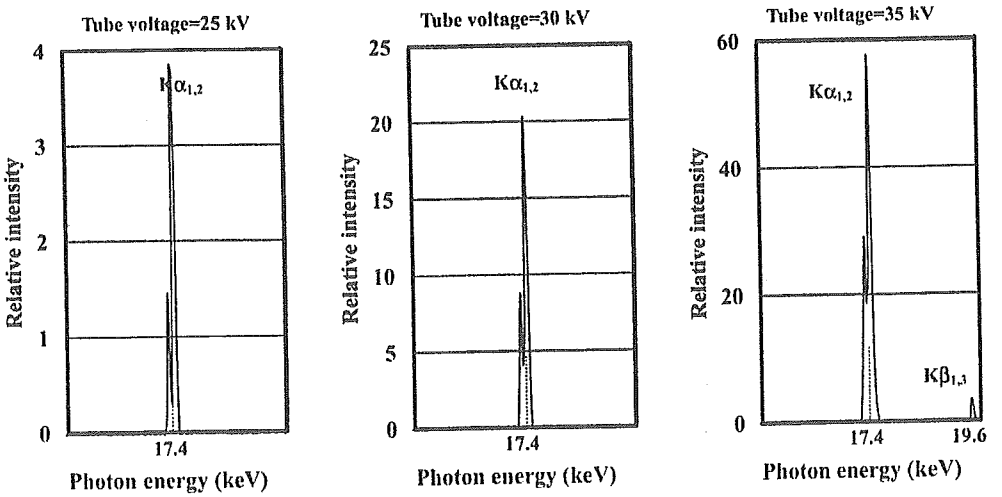

Fig. 12. X-ray spectra from the molybdenum target.

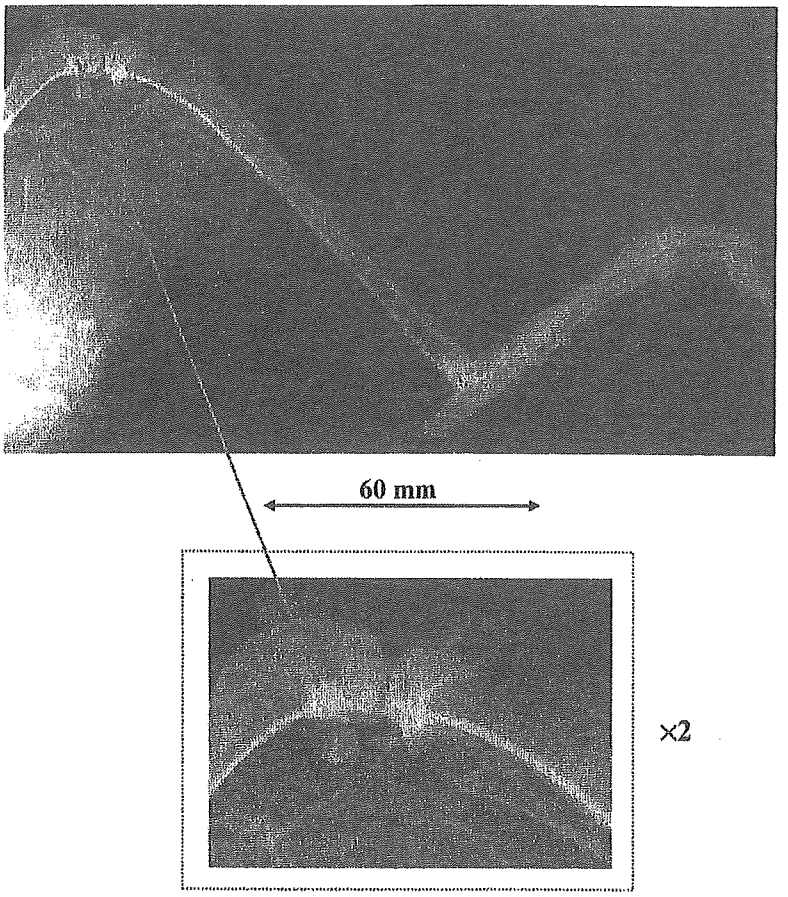

Fig. 13. Angiogram of a rabbit thigh.

6. セリウムX線装置とヨウ素Kエッジ造影

コーンビームを用いてシンクロトロンと同等のヨウ素Kエッジ造影効果を得るため，Kα線の平均フォトンエネルギーが 34.6 keV のセリウムX線管[24,25]を製作し，高電圧電源と接続してポータブルX線装置を製作した。装置はメインコントローラーとコッククロフト・ウォルトン回路を組み込んだX線管ユニットからなる。メインコントローラーでは管電圧，管電流，そしてX線照射時間を調整できるが，コントローラーに接続したパソコンを用いても遠隔から制御できる。X線管の陽極は接地され，陰極に負の高電圧が印加される。X線管の焦点径は約 1 mm で，フィルター無し，そして硫酸バリウムフィルターを付けた場合のX線強度は，線源から 1.0 m の位置で，それぞれ 209 および 16.8 μGy/s であった。このとき，管電圧と電流は，それぞれ 60 kV と 0.5 mA であった。

Fig. 14 はセリウムX線管からのX線スペクトルで，管電圧が 60 kV でのものである。図のようにシャープなK線が得られ，硫酸バリウムフィルターの挿入によって Kβ線強度は著しく減弱した。ヨウ素マイクロスフェアーを用いた犬の心臓の造影では，厚さ 100 mm の水ファントムを用いた場合にでも，血管のコントラストはほとんど変化しなかった（Fig. 15）。

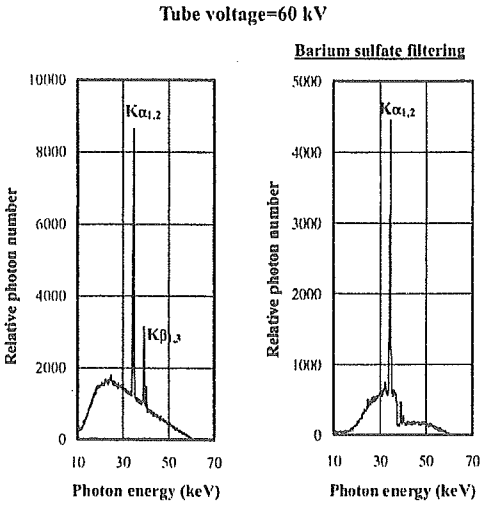

Fig. 14. X-ray spectra from a cerium target.

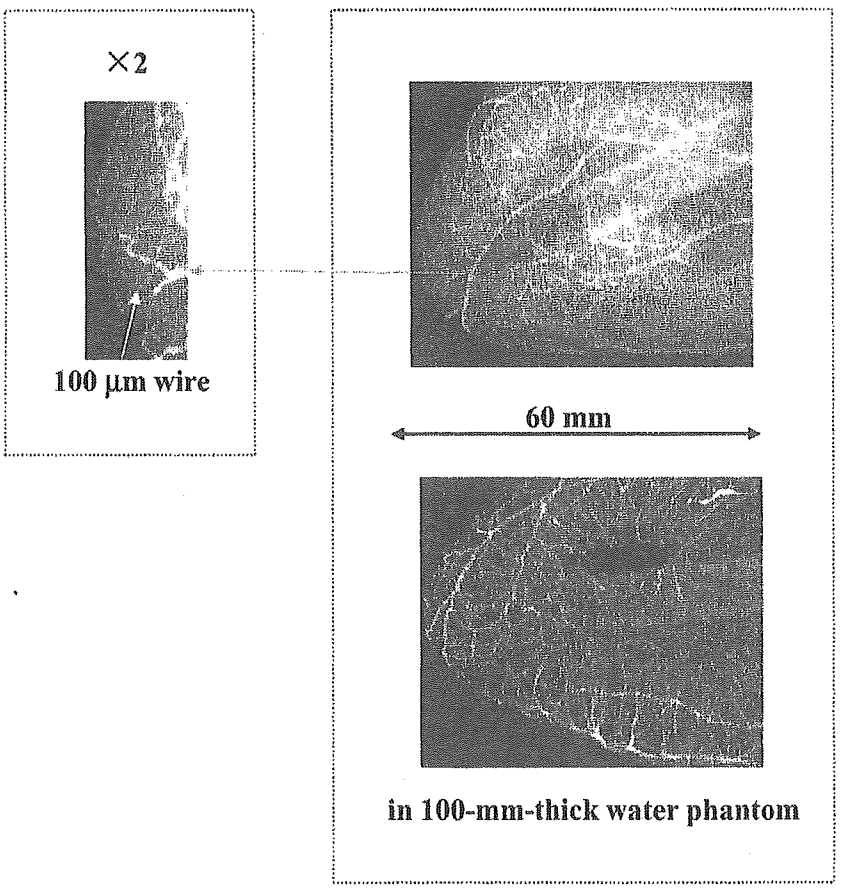

Fig. 15. Angiograms of a dog heart.