Table 1. MK4 Cancer Research (Human)

Study	Citation	Indications	Design	Volunteers	MK4 Dose	Duration	Outcomes	Adverse Events
Myelodysplastic Syndrome (MDS) and Acute Myeloid Leukemia (AML)								
Akiyama and Miyazawa	*Leukemia Research*. 2010; 34(9): 1151-1157.	MDS	Multicenter, open-labeled, single-arm, prospective phase II clinical trial	n = 38 (20 male, 18 female); median age 65 years	45 mg/day orally or MK4 + Vitamin D	16 weeks	13% in the MK4 group and 30% in the MK4 + Vitamin D group improved as defined by IWG-2000 criteria.	1 case of grade I nausea, 1 of grade I lower abdominal pain, and 1 of grade I skin rash
Takami and Asakura	*Annals of Hematology*. 2002; 81(1):16-9	MDS	Prospective, randomized trial	n = 18 (9 male, 9 female); mean age 72 years	45 mg/day orally	38 months	56% (5/9) in MK4 group improved as defined by IWG-2000 criteria	No signs of toxicity or progression to acute leukemia were observed.
Miyazawa and Nishimaki	*Leukemia*. 2000; Jun;14(6):1156-7	MDS and Post-MDS AML	Retrospec-tive, question-naire survey at 11 different institutions	n = 47 (30 male, 17 female); median age 66 years	20-135 mg /day orally, and 10-50 mg /day intravenous-ly	Not stated.	MK4 alone: 44.4% (4/9) of RAEB-T and Post-MDS AML showed hematological improvement.	None

Study	Citation	Indications	Design	Volunteers	MK4 Dose	Duration	Outcomes	Adverse Events
Takami and Nakao	*International Journal of Haemato-logy*.1999;69 (1):24-6	MDS with refractive anemia (RA)	Case report	80-year-old woman	45 mg/day orally	14 months	Woman's health improved and transfusions were no longer needed. When MK4 was stopped, her blood results worsened, but improved again when MK4 (45 mg/d) was restarted.	Not reported
Yaguchi and Miyazawa	*Leukemia*. 1999;13(1):144-5	RAEB-T with myelofibro-sis (MDS/AML)	Case report	65-year-old male	90 mg/day orally, reduced to 45 mg/day orally after hematologic al response	10 months (6 weeks at 90 mg/day orally and the remaind er at 45 mg/day orally	Peripheral blast cell count significantly decreased, platelet count normalized	None

Study	Citation	Indications	Design	Volunteers	MK4 Dose	Duration	Outcomes	Adverse Events
Liver Cancer (Hepatocellular Carcinoma)								
Ishizuka and Kubota	*Anticancer Research*. 2012; 32(12):5415-20	Hepatocellu-lar carcinoma following hepatic-tomy surgery	Prospective, randomized controlled trial	n = 101 (78 men, 23 women); mean age approx. 67 years.	MK4 45 mg/day	17-68 months (median = 34 months)	The researchers concluded that MK4 had a "suppressive effect on HCC recurrence between 15 months and 4 years after surgical resection, especially in patients with a normal preoperative DCP level" and recommend that MK4 "should be considered as an adjuvant agent for prevention of HCC recurrences after surgery"	

Study	Citation	Indications	Design	Volunteers	MK4 Dose	Duration	Outcomes	Adverse Events
Yoshiji and Noguchi	*World Journal of Gastroenterology*. 2007; 13(23):3259-61	Hepatocellular carcinoma with liver cirrhosis, hepatitis C	Case report	n = 1 (woman); 66 years old	MK4 45 mg/day orally plus perindopril 4 mg/day orally	5 years	Blood markers--AFP and lectin-reactive alpha-fetal protein (AFP-L3) normalized after 1 year. After 15 months of treatment, the hepatic nodule disappeared. MK4 and ACE-I was continued, and neither AFP nor AFP-L3 levels increased again. This combination was continued, and no HCC developed.	None

Study	Citation	Indications	Design	Volunteers	MK4 Dose	Duration	Outcomes	Adverse Events
Kakizaki and Sohara	*Journal of Gastroenter ol-ogy and Hepatology*. 2007; 22(4):518-22	Hepatocellu-lar carcinoma with Hepatitis C	Randomized , controlled	n = 60 (35 men, 25 women); mean age 69 years who had undergone radiofreque n-cy ablation	MK4 45 mg/day orally or no MK4	36 months	After 36 months, 38.8% of volunteers taking MK4 were "recurrence-free" compared to only 9% of those not taking MK4. At 36 months, 77.5% of those taking MK4 were still alive versus 66.4% of those who did not take MK4.	None

Study	Citation	Indications	Design	Volunteers	MK4 Dose	Duration	Outcomes	Adverse Events
Mizuta and Ozaki	*Cancer*. 2006; 106(4):867-72	Hepatocellu-lar carcinoma and Hepatitis C	Randomized , controlled	n = 61 (41 men, 20 women); mean age 64 years	MK4 45 mg/day orally or no MK4	36 months	Cumulative survival for volunteers who received MK4 were 100% at 12 months, 96.6% at 24 months, and 87.0% at 36 months; and the corresponding survival rates for patients in the control group were 96.4%, 80.9%, and 64.0%. After 3 years, 23% more people taking MK4 were alive compared to those not taking MK4.	None

Study	Citation	Indications	Design	Volunteers	MK4 Dose	Duration	Outcomes	Adverse Events
Yoshiji and Noguchi	*Journal of Hepatology*. 2005; 42(5):687-93	Hepatocellu-lar carcinoma with Hepatitis C	Phase I: Randomized , controlled	n = 60 (52 men, 32 women); mean age 50 years. All had undergone percutaneou s radiofreque n-cy ablation (RFA), considered "curative therapy"	Group A: no intervention control Group B: MK4 45 mg/day orally plus perindopril 4 mg/day orally	48 months		None

Study	Citation	Indications	Design	Volunteers	MK4 Dose	Duration	Outcomes	Adverse Events
Habu and Shiomi	*Journal of the American Medical Association* (JAMA). 2004; 292(3):358-361	Liver cirrhosis becoming liver cancer with Hepatitis C	Randomized, controlled	n = 43 (all women); mean age 60 years	MK4 45 mg/day orally or no MK4	8 years	After 8 years only two volunteers taking MK4 developed liver cancer compared to nine volunteers in the group not taking MK4. The researchers concluded that MK4 decreased the chance of developing liver cancer in this study by 80%.	None

Study	Citation	Indications	Design	Volunteers	MK4 Dose	Duration	Outcomes	Adverse Events
Nouso and Uematsu	*World Journal of Gastroenterol-ogy.* 2005; 11(42):6722-4	Hepatocellu-lar carcinoma with Hepatitis C and diabetes mellitus	Case report	n = 1 (man); 85 years old who declined interventional therapies or angiography "because of the patient's advanced age and advanced stage of HCC"	MK4 45 mg/day orally	2 months	Blood markers (serum alpha-fetal protein, AFP) and des-gamma-carboxy-prothrombin (DCP) normalized after 3 months. Tumor decreased from 13.3 cm to 5.5 cm after 5 months. Ultrasound demonstrated the tumor regressed and the margin of the tumor became obscure. After 22 months after initiating MK4 there were no signs of recurrence observed and tumor markers remained within the normal limits.	Not reported

Study	Citation	Indications	Design	Volunteers	MK4 Dose	Duration	Outcomes	Adverse Events
Jancin	*Family Practice News*. 2002; 32(14):16(11)	Hepatocellu-lar carcinoma	Randomized, controlled	n = 121 who had received conventional therapy with percutaneous tumor ablation and/or transcatheter arterial embolization.	MK4 45 mg/day orally or no MK4	24 months	Rate of portal vein invasion at 12 months of follow-up was 2% in those taking MK4 and 23% in controls. At 2 years, portal vein invasion was present in 23% of the MK4 group and 47% of controls. Additionally, one-year survival in patients treated with MK4 was 76%, compared with 66% in controls. Two-year survival was 66% in the MK4 group and just 28% in controls.	Not reported
		Hepatocellu-lar carcinoma with Hepatitis C	Phase II: Randomized, controlled	n = 40; mean age 61 years	Group A: MK4 45 mg/day orally Group B: perindopril 4 mg/day orally	36 months	Cumulative recurrence after four years was 36%, meaning 64% exhibited normal test results.	None

Study	Citation	Indications	Design	Volunteers	MK4 Dose	Duration	Outcomes	Adverse Events
Acute Promyelocytic Leukemia								
Fujita and Tomiyama	British Journal of *Haematology*. 1998. 103(2): 584.	Acute promyelocytic leukemia refractive to conventional chemo-therapy	Case report	n = 1 (woman); 72 years old with t(15;17) translocation	MK4 20 mg/day orally plus all trans retinoic acid	2 months	After 7 d promyelocytes disappeared from peripheral blood. Two months later, bone marrow aspiration revealed complete remission. In addition, genetic abnormalities (PML/RARa fusion transcript and t(15;17) translocation) disappeared.	Not reported

Table 2. MK4 Cancer Research (in vitro)

Study	Citation	Model	Cell line	Control	MK4 Dose	Duration	Outcomes
Leukemia Cell Lines							
Tsujioka and Miura	*Haematologica*. 2006; 91(5):613-9.	Myeloma, Promyelocytic leukemia, Follicular lymphoma, T-cell acute leukemia, Adult T-cell lymphoma/leu-kemia, histiocytic leukemia, Burkitt's lymphoma	U266 and RPMI8226, KMM-1, KMS-11, KMS-12PE, KMS-12BM. KMS-20, KMS-24, KMS-26, KMS-27, KMS-28PE, KMS-34, SU-DHL-4, CEM, Molt4 and Jurkat, MT-1		0 to 30 μM	Up to 72 hours	↑Apoptosis ↑Mitochondrial apoptotic pathway ↑Caspase-3 ↓ Bcl-XL/XS expression

Study	Citation	Model	Cell line	Control	MK4 Dose	Duration	Outcomes
Miyazawa and Yaguchi	*Leukemia*. 2001; 15(7):1111-7.	Leukemia	BCL-2 overexpressed HL-60 leukemic cell line by bcl-2 gene transfection (HL-60-bcl-2)	HL-60-neo cells	0.1 to 50 μM	72 hours	↑Apoptosis in HL-60-neo but not HL-60-bcl-2 ↑Differentiation ↑G0/G1 arrest in HL-60-bcl-2 that were resistant to MK4-inducing apoptosis, and also HL-60-neo cells ↓ Cell growth at 5 μM in HL-60-bcl-2 cells ↑ p27KIP1 (a CDK inhibitor) in HL-60-neo cells ↑Monocytic differentiation in HL-60-neo cells evidenced by increased CD-14, CD-15 and CD16 antigen expression IC50 6 μM for HL-60-neo cells 14 μM for HL-60-bcl-2 cells
					10 mm	24 hours	↑Depolarization of the mitochondrial membrane potential in HL-60-neo cells ↑Caspase-3 in HL-60 neo but not HL-60-bcl-2

Study	Citation	Model	Cell line	Control	MK4 Dose	Duration	Outcomes
Nishimaki and Miyazawa	*Leukemia*. 1999; 13(9):1399-405.	MDS in blastic transformation (MDS becoming AML)	Established from a 73-year-old female with MDS, RAEB-T, with chromosomal anomalies chromosomal anomaly including -4, 5q-, -7, 20q-		10 µM	72 hours	↓ BCL-2 ↑ BAX ↑ Caspase-3

Study	Citation	Model	Cell line	Control	MK4 Dose	Duration	Outcomes
Yaguchi and Miyazawa	*Leukemia*. 1997; 11(6):779-87.	MDS and Post-MDS AML, CML, Promyelocytic leukemia	Leukemia cells freshly isolated from bone marrow or peripheral blood samples from 12 patients diagnosed with AML and MDS based on FAB criteria. Patients included four with AML (two M2 and two M3), two with CML in blastic crisis, three with MDS (one RAEB, one RAEB-T and on CMML), and three with post-MDS AML.	MDS92, an IL-3 dependent myeloid cell line established from a patient with MDS NB4, a cell line established from a patient with promyelocytic leukemia, and MDS92, an IL-3-dependent myeloid cell line from a patient with MDS	10 μM	48 hours	Apoptosis observed in cells from all 12 patients. IC50 ≈ 10 μM Exposure selectively eliminated the leukemic cell populations

Study	Citation	Model	Cell line	Control	MK4 Dose	Duration	Outcomes
Liver Cancer (Hepatocellular Carcinoma)							
Azuma and Urano	*Endocrine Journal*. 2009; 56(7):843-9.	In vitro	HepG2, HuH7 as HCC cell lines. LS180, a colorectal cancer cell line, as a positive control		10 µM	24 hours	↓ Steroid and xenobiotic receptor (SXR), also called pregnane X receptor (PXR) ↓Cell motility, an indicator of decreased potential of cancer cells to metastasize (spread to other parts of the body)
Ide and Zhang	*Oncology Reports*. 2009; 22(3): 599-604	In vitro	HepG2, HuH7, HLE		0, 10^{-6}, $10^{-5}$and 10^{-4} M	48 hours	↓ Matrix metalloproteinases (MMP) expression in a dose-dependent manner ↓ Protein kinase C (PKC) activity ↓ MMP gene promoter activity ↓ NFκB MK4 activity was dose-dependent

Study	Citation	Model	Cell line	Control	MK4 Dose	Duration	Outcomes
Ozaki and Zang	*Clinical Cancer Research*. 2007; 13(7):2236-45.	In vitro	HepG2, Hep3B and Huh7 human HCC cell line	No MK4	10-6 M, 10-5 M and 10-4 M	48 hours	↓HCC cell growth in a dose-dependent manner ↓DNA synthesis in HCC cells ↑ G1 cell cycle arrest ↓ Cyclin D1 expression at the mRNA and protein level ↑p21 and p27 expression ↓ −1745CD1Luc, −964CD1Luc, −964AP-1mutCD1Luc, and −66CD1Luc promoters ↓ NFκB expression and DNA binding

Study	Citation	Model	Cell line	Control	MK4 Dose	Duration	Outcomes
Otsuka and Kato	*Hepatology*. 2004; 40(1):243-51.	In vitro	HepG2 and PRF/PLC/5, murine embryonic fibroblasts (NIH3T3) and a human primary hepatocyte cell culture (ACBRI)	No MK4	0 µM, 30 µM, 50 µM and 80 µM	5 days	↑protein kinase A (PKA) ↓cell growth MK4 did not adversely effect "primary hepatocytes" (non-cancerous cells) ↓cancer cell invasion by increasing PKA activity ↑DNA transcription in 3.4% of 10,000 genes evaluated ↑AP-2–, USF-1–, and CREB-related transcriptional activities ↓RhoA activation ↓ DCC production IC_{50}= 45 µM
		Animal	Athymic nude mice inoculated subcutaneously with 1x107 PRF/PLC/5 cells, then tumors allowed to develop for two weeks.	Ethanol	20 mg/kg/d MK4 by mouth	6 weeks	Four of the five ethanol-treated mice developed large visible tumors. While three of the four MK4-treated mice also developed visible tumors, the tumors were smaller than those in ethanol-treated mice. Also, body weight decreased less in the animals fed MK4.

Study	Citation	Model	Cell line	Control	MK4 Dose	Duration	Outcomes
Cholangiocellular Carcinoma (CCC)							
Enomoto and Tsuchida	*International Journal of Molecular Medicine*. 2007; 20(6):801-8.	In vitro	Cholangiocellular carcinoma (CCC) cell lines TFK-1, MEC and HuCC-T1	HL-60 leukemic cells	0.1, 0.5, 1, 2, 5, 10, 20, 50 and 100 μM	96 hrs	Dose-dependent cell growth inhibition. ↓CCC cell growth MK4 induced autophagy in CCC cells IC_{50} = 7.3-10.7 for the three cell lines.