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# Adipogenesis in a Myeloid Supporting Bone Marrow Stromal Cell Line

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**Abstract** The bone marrow stroma contains pre-adipocyte cells which are part of the hemopoietic microenvironment. Cloned stromal cell lines differ both in their ability to support myeloid and lymphoid development and in their ability to undergo adipocyte differentiation in vitro. These processes have been examined in the +/+2.4 murine stromal cell line and compared to other stromal and pre-adipocyte cell lines. In long-term cultures, the +/+2.4 stromal cells support myeloid cell growth, consistent with their expression of macrophage-colony stimulating factor mRNA. However, despite the presence of mRNA for the lymphoid supportive cytokines interleukins 6 and 7, +/+2.4 cells failed to support stromal cell dependent B lineage lymphoid cells in vitro, suggesting that these stromal cells exhibit only a myelopoietic support function. The +/+2.4 cells differentiate into adipocytes spontaneously when cultured in 10% fetal bovine serum. The process of adipogenesis can be accelerated by a number of agonists based on morphologic and gene marker criteria. Following induction with hydrocortisone, methylisobutylxanthine, indomethacin, and insulin in combination, a time dependent increase in the steady state mRNA and enzyme activity levels of the following adipocyte specific genes was observed: adipocyte P2, adipsin, CAAT/enhancer binding protein, and lipoprotein lipase. In contrast, adipogenesis was accompanied by a slight decrease in the signal intensity of the macrophage-colony stimulating factor mRNA level, similar to that which has been reported in other bone marrow stromal cell lines. These data demonstrate that although the lympho-hematopoietic support function of pre-adipocyte bone marrow stromal cell lines is heterogeneous, they share a common mechanism of adipogenesis. 

**Key words:** adipocyte, differentiation, CCAAT/enhancer binding protein, lipoprotein lipase, adipsin, macrophage colony stimulating factor

The bone marrow stroma provides the lympho-hemopoietic microenvironment in adult vertebrates necessary for the normal production of circulating blood cells [Kincade et al., 1988; Weiss and Sakai, 1984]. Stromal cells accomplish this task through the release of cytokines which regulate the proliferation and maturation of pluripotent hematopoietic stem cells. Among these cytokines are macrophage-colony stimulating factor (M-CSF), regulating macrophage development, interleukin-7 (IL-7), enhancing lymphoid proliferation, and interleukin-6 (IL-6), which can synergize with other cytokines. Morphologically, the stroma is heterogeneous, consisting of fibroblast/endothelial cells, adipocytes, and macrophages. In mammals, increasing age is associated with the accumulation of adipose tissue within the bone marrow cavity [Bathija et al., 1979; Custer and Ahlfeldt, 1932; Hartsock et al., 1965; Ricci et al., 1990; Rozman et al., 1989]. The role of bone marrow fat remains controversial [Gimble, 1990; Tavassoli, 1984]. Unlike extramedullary adipose tissue, the bone marrow adipocytes do not respond to starvation with increased lipolysis [Bathija et al., 1979]. However, stresses such as anemia rapidly deplete bone marrow adipose reserves in association with elevated erythropoiesis [Bathija et al., 1978; Brookoff and Weiss, 1982; Tavassoli et al., 1974]. Following advances in long-term bone marrow culture methods, a number of stromal cell lines have been cloned and characterized with respect to their ability to support hemo-

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poiesis in vitro, their production of specific cytokines, and their ability to differentiate into adipocytes [Gimble, 1990; Zipori, 1989]. Not unexpectedly, these lines have proven to be heterogeneous. While some of these stromal cells only support myelopoiesis in vitro, others are distinguished by their ability to support B-lymphopoiesis as well. Nevertheless, members of both categories of stromal cells have been observed to differentiate into adipocytes.

Earlier work documented the process of adipogenesis in the B-lymphopoietic supporting BMS2 murine stromal cell line [Gimble et al., 1989, 1990]. The current paper examines adipogenesis at the cellular and molecular levels in the myelopoietic supporting +/+2.4 murine stromal cell line +/+2.4 [Anklesaria et al., 1987]. These cells were originally derived by limiting dilution techniques from the adherent layer of murine long-term bone marrow cultures [Anklesaria et al., 1987]. The response of +/+2.4 cells to adipogenic agonists and the kinetics of adipocyte marker gene expression are determined. These data provide support for a generalized mechanism for adipogenesis within bone marrow stromal cell lines, regardless of their hemopoietic support potential.

## MATERIALS AND METHODS

### Cell Culture

The +/+2.4 cells [Anklesaria et al., 1987] were cultured in RPMI 1640 medium supplemented with 10% fetal bovine serum (Whitaker, Walkersville, MD), 1 mM sodium pyruvate, 100 units/ml penicillin, 100 mcg/ml streptomycin, and 50 mcM 2-mercaptoethanol. Inductions were performed in the same medium with the addition of 5% calf serum (Hyclone, Logan, UT). The following agents were employed at the indicated concentrations. All reagents were purchased from Sigma Chemical Co. (St. Louis, MO) unless otherwise indicated, and used at the following concentrations: hydrocortisone, $5 \times 10^{-7}$ M (Elkin-Sinn, Cherry Hill, NJ); dexamethasone, $5 \times 10^{-7}$ M; insulin, 5 mcg/ml; indomethacin, 0.06 mM; methylisobutylxanthine (MIBX), 0.5 mM. Confluent, quiescent cultures of +/+2.4 cells in 35 mm plates (Falcon or Corning) were treated for a period of 60 h as follows: no additions (control); glucocorticoid alone (dexamethasone or hydrocortisone); glucocorticoid, MIBX, and insulin together; or glucocorticoid, MIBX, insulin, and indomethacin together. At the end of the induction period, these agents were removed and the cells maintained in the 10% fetal bovine serum/RPMI 1640 media described above. Cultures were fed routinely every 3 days with the replacement of 50% of the culture medium. The degree of adipocyte differentiation was determined in five mid power (×20) fields using a Zeiss IM-35 phase contrast microscope.

### Methylcellulose Assays

Methylcellulose co-cultures of the stromal cell dependent B lineage lymphoid cell lines 1A9 and 2E8 with either +/+2.4 or BMS2 stromal cells were performed using $5 \times 10^4$ stromal cells and $10^4$ lymphoid cells per 35 mm dish as previously described [Ishihara et al., 1991].

### RNA Purification and Northern Blot Analysis

Total RNA was prepared from cultures according to the method of Chomczynski and Sacchi [1987] as previously described [Gimble et al., 1989]. Northern blots were prepared with 10–20 mcg of total RNA per lane according to the method of Thomas [Gimble et al., 1989; Thomas, 1980].

### Random Priming and Hybridization

The cDNA probes utilized were radiolabeled with [alpha$^{32}$-P] dCTP by the random primer method of Feingold and Vogelstein [1983]. Hybridizations were performed as described [Gimble et al., 1989]. Probes were obtained courtesy of the following individuals: aP2, Howard Green, Harvard University [Spiegelman et al., 1983]; adipsin, William Wilkison, Dana Farber Cancer Center [Wilkison et al., 1990]; CHO-B, Randall Wall, UCLA [Harpold et al., 1979]; murine C/EBP, K. Xanthopoulus, Karolinska Institute, and Sven Enerback, University of Goteborg [Landschulz et al., 1988]; M-CSF (pGEM2M-CSF10, culture collection #CMCC 2760), Peter Ralph, Cetus Corp.

### Lipoprotein Lipase Assay

The medium from adipocyte cultures was replaced with fresh medium containing 10 units/ml heparin. Cultures were incubated for 1 h at 37°C and the medium immediately harvested and stored at −70°C. The LPL activity was determined according to the method of Nilsson-Ehle and Schotz [1976] as described [Gimble et al., 1989].

### Cloning of the Lipoprotein Lipase cDNA

A murine specific probe for the 5′ end of the LPL cDNA was cloned according to a modifica-

tion of the rapid amplification of cDNA ends (RACE) protocol of Frohman et al. [Cooper and Isola, 1990; Frohman et al., 1988]. A specific primer complementary to the coding sequence was synthesized based on bp 555–600 of the published sequence of Kirchgessner et al. [1987]. This primer, together with commercially available primers complementary to the cloning sites of lambda gt11, were used to screen a BMS2 adipocyte cDNA library (purchased from Clontech Laboratories, Palo Alto, CA) prepared in lambda gt11 with oligo dT and random hexamers. Polymerase chain reactions were carried out between the LPL specific and the lambda gt11 based primers using approximately $1 \times 10^7$ plaque forming units as template. The template was initially treated using a Cetus-Perkin-Elmer Thermal Cycler for 5 min at each of the following temperatures prior to the addition of the primers, deoxynucleotides, and Taq polymerase enzyme (Promega, Madison, WI): 72°C, 94°C, 55°C. Forty cycles were performed at 94°C (1 min), 55°C (2 min), and 72°C (3 min). The products were purified using the Geneclean method (Bio101, LaJolla, CA) and digested with the restriction enzymes EcoRI and BamHI. The fragments were subcloned into the polylinker of Bluescript SKII (Stratagene, La Jolla, CA) and insert size determined based on PCR reaction with vector specific primers. Plasmids with the largest inserts (711 bp) were selected and sequenced according to the dideoxy method [Sanger et al., 1977] using the Sequenase enzyme (US Biochemicals, Cleveland, OH) [Tabor and Richardson, 1987]. The sequence of the cDNA was in agreement with those of the first exon of the murine LPL [Hua et al., 1991; Zechner et al., 1991] and of the previously published LPL cDNA [Kirchgessner et al., 1987; Semenkovich et al., 1989]. The sequence has been entered into the GenBank (accession #M65258) and the plasmid designated OMRF3405 has been placed on deposit with the American Tissue Culture Collection, Rockville, MD (catalog #63117). The BamHI/EcoRI insert was radiolabeled as described above and used in Northern blot hybridizations.

### Computer Analysis

Sequence analysis was performed using the University of Wisconsin Genetics Computer Group Software Programs [Devereux et al., 1984].

## RESULTS

### Spontaneous Differentiation and Lympho-Hematopoietic Support Function of +/+2.4 Cells

Cultures of +/+2.4 cells were noted to undergo spontaneous differentiation into adipocytes in the presence of fetal bovine serum. To investigate this process, total RNA from preadipocyte and adipocyte +/+2.4 cells was compared on Northern blots to that of two well-characterized adipocyte models, BMS2 stromal cells and 3T3-L1 fibroblasts (Fig. 1). The adipocyte marker genes adipocyte P2 (aP2) and adipsin were not detected in total RNA from any pre-adipocyte cells. These genes were induced in +/+2.4 adipocytes in a manner similar to the other adipocyte models. Macrophage-colony stimulating factor (M-CSF) mRNA was detectable in both pre-adipocyte and adipocyte populations in all cell lines. Consistent with this, the +/+2.4 cell lines exhibited myelopoietic support in long term bone marrow cultures (Fig. 2C), as earlier described [Anklesaria et al., 1987]. Under Dexter (myeloid supporting) culture conditions, these stromal cells provided a microenvironment suitable for the proliferation of myeloid cells. The expression of mRNA for other less abundant cytokines was examined by PCR. Specific oligonucleotide probes from different exons of interleukin-6 and interleukin-7 were used to amplify the cDNA from pre-adipocyte and adipocyte total RNAs (data not shown). As a positive control, the BMS2 cell line was used. The products were examined on southern blots hybridized with IL-6 or IL-7 specific probes. Signals of appropriately sized fragments for a cDNA template were detected with both interleukin probes in preadipocyte and adipocyte +/+2.4 cells, suggesting that these cytokines are expressed at the mRNA level. However, despite the expression of IL-7 mRNA, the +/+2.4 cells failed to support the proliferation of the IL-7 dependent B lymphoid cell line (2E8) in methylcellulose assays (Table I), suggesting that they do not express the protein itself or lack other necessary cell surface molecules required for B lymphopoietic support function. A second stromal cell dependent B lineage lymphoid cell (1A9) also failed to proliferate in culture with the +/+2.4 cells (Table I). In parallel experiments, both lymphoid lines were supported by BMS2 cells. These data indicate that +/+2.4 cells act as stromal cells for myelopoiesis only.

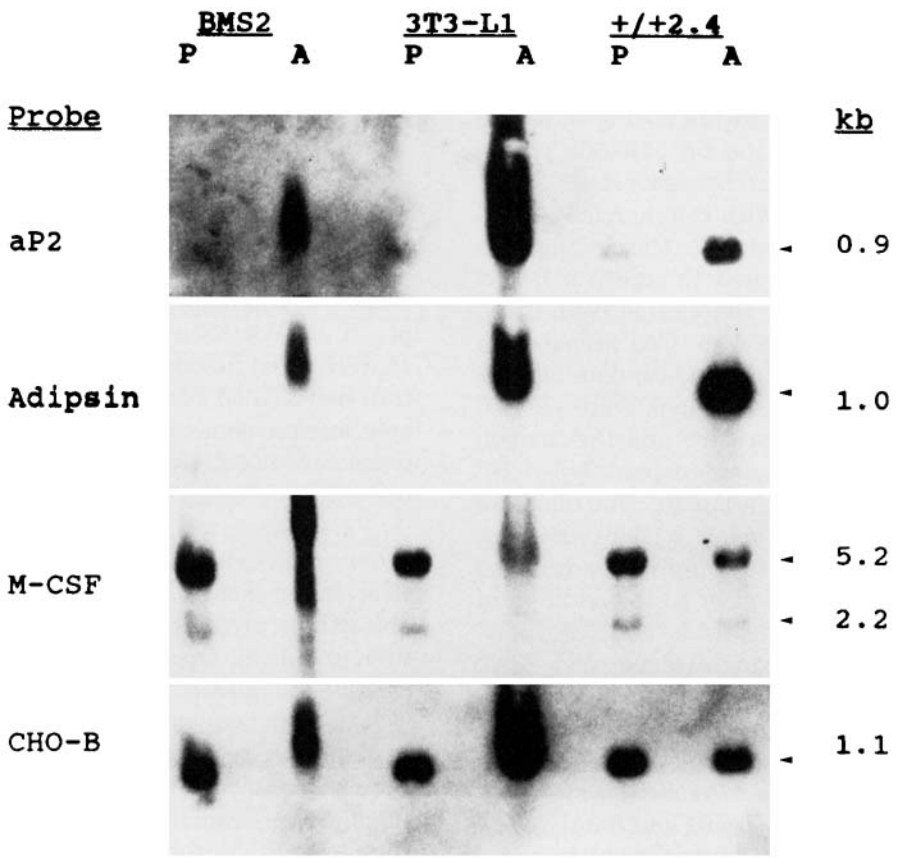



**Fig. 1.** Comparison of gene expression in pre-adipocyte and adipocyte cells. Northern blot analysis. Total RNA from preadipocyte (P) and adipocyte (A) BMS2, 3T3-L1, and +/+2.4 cells was harvested as described in Materials and Methods and examined on Northern blots prepared with approximately 10 mcg total RNA/ lane. The +/+2.4 adipocytes had been allowed to differentiate spontaneously. Identical blots were successively hybridized with cDNA probes for adipsin, aP2, MCSF, and CHO-B, a control housekeeping gene which is relatively unaffected by adipocyte differentiation. The transcript sizes (kb) are indicated.

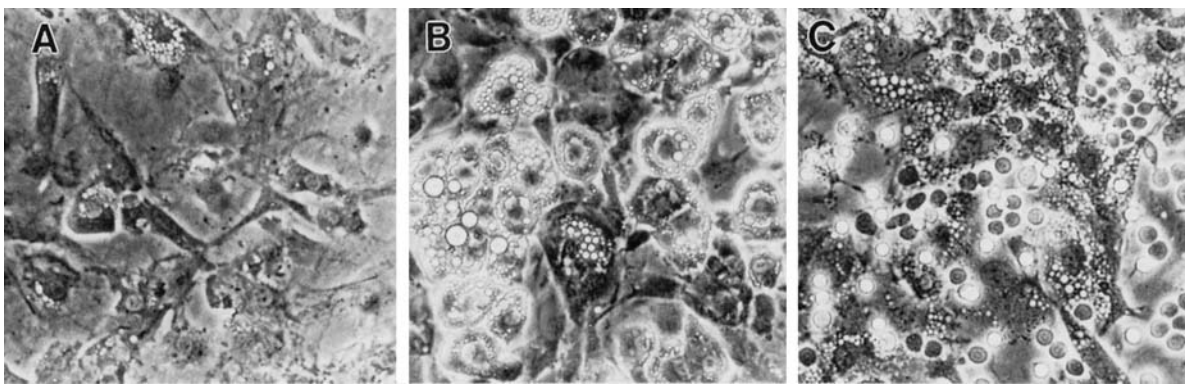



**Fig. 2.** Photomicrographs of +/+2.4 cultures. **A:** Phase contrast photomicrograph of uninduced pre-adipocyte +/+2.4 culture at ×20 magnification on a Zeiss IM35 microscope. **B:** Phase contrast photomicrograph of induced (MIBX/hydrocortisone/indomethacin/insulin) +/+2.4 cells at ×20 magnification. **C:** Phase contrast photomicrograph of myelopoietic Dexter cultures prepared with +/+2.4 stromal cells and nylon wool passage (stromal cell depleted) Balb/c murine bone marrow cells at ×32 magnification.

**TABLE I. Methylcellulose Assays of B Lineage Lymphoid Cell Support**

| | B lineage lymphoid cells[a] | |
|---|---|---|
| | 2E8 | 1A9 |
| Stromal cells[b] | | |
| BMS2 | 330 ± 21 | 409 ± 82 |
| +/+2.4 | 0 | 0 |

[a]Lymphoid cells were plated at $10^4$ cells per 35 mm dish in 2.1% methylcellulose. Colony counts were determined after 7 days in culture. Values are the mean +/− the standard deviation from 3 separate experiments.
[b]Stromal cells were plated at $5 \times 10^4$ cells per 35 mm dish.

### Response of +/+2.4 Cells to Adipogenic Agonists

Confluent cultures of quiescent +/+2.4 cells were induced with a combination of adipogenic agonists and the number of phenotypical adipocytes based on phase contrast microscopy monitored over 6 days (Figs. 2A,B, 3A,B). The cells were cultured in the absence of exogenous agents (control, C) or in the presence of glucocorticoids (dexamethasone, D, or hydrocortisone, H) without or with methylisobutylxanthine (M), a phosphodiesterase inhibitor, and indomethacin (I), a prostaglandin synthesis inhibitor. Photomicrographs of pre-adipocytes and MHI-induced adipocytes are presented in Figure 2A,B. In comparison to control cultures, the addition of glucocorticoids alone did not significantly increase the number of cells exhibiting an adipocyte morphology (Fig. 3A,B). However, the combinations of glucocorticoids, insulin, and MIBX without (MH, MD) or with indomethacin (MHI, MDI) accelerated the rate of adipocyte differentiation. By day 5, the number of phenotypic adipocytes under these conditions was significantly greater than the untreated controls.

After 6 days in culture, the cells in the experiment were harvested for total RNA and the steady state mRNA levels of adipocyte specific gene markers determined on Northern blots (Fig. 3C). The following genes were examined: aP2, adipsin, CAAT/enhancer binding protein (C/EBP), and lipoprotein lipase (LPL). The constitutively expressed housekeeping gene CHO-B was used as a control for relatively equal loading between lanes. The pattern of gene expression correlated directly with the morphologic analysis. The signal intensity of each mRNA was relatively lower in the untreated controls (C) and cells treated with glucocorticoid alone (H, D) when compared to the cells induced in the presence of MIBX without or with indomethacin (MH, MD, MHI, MDI). Together, these data indicate that the adipogenic agonists accelerate the rate of differentiation in +/+2.4 cells.

### Expression Kinetics of Adipocyte Differentiation Marker Genes

The kinetics of adipocyte specific gene steady state mRNA levels were examined on Northern blots (Fig. 4). Total RNA was harvested daily from +/+2.4 cells prior to and after induction with the combination of hydrocortisone, methylisobutylxanthine, indomethacin, and insulin. Prior to induction, there was no detectable signal for any of the genes examined. However, within 2–3 days of induction, mRNA for C/EBP, aP2, and adipsin was present. Maximum levels were achieved by days 6–8 post-induction. In contrast, the level of M-CSF mRNA declined slightly over the same period.

### Cloning of Lipoprotein Lipase cDNA Probe and Kinetics of Lipoprotein Lipase Expression

A probe for the murine LPL cDNA was cloned from a BMS2 adipocyte library using a polymerase chain reaction based method. The sequence of the 711 bp insert was determined (Genbank accession #M65258) and its 5′ end found to be identical to the genomic sequence of the untranslated region of the LPL first exon [Hua et al., 1991; Zechner et al., 1991]. This probe was employed in kinetic analysis of LPL expression in differentiating +/+2.4 cells. Induced cells were harvested daily for their heparin releasable LPL enzyme activity and total RNA. Prior to induction, detectable levels of LPL mRNA were present in +/+2.4 cells (Fig. 5A). The estimated size of the murine LPL mRNAs (4.1 kb, 4.5 kb) was consistent with the recently published full-length murine LPL genomic sequence [Zechner et al., 1991]. The full-length murine cDNA is at least 3.97 kb in size and is approximately 400 bp larger than its human counterpart [Zechner et al., 1991]. The steady state mRNA levels of LPL increased within 1 day of induction and reached maximum levels between days 6 and 9 post-induction. The mRNA levels paralleled the temporal increase of LPL enzyme activity (Fig. 5B). Heparin releasable LPL activity increased five to sixfold over the induction period, reaching maximum levels at day 8 to 9 post-induction.

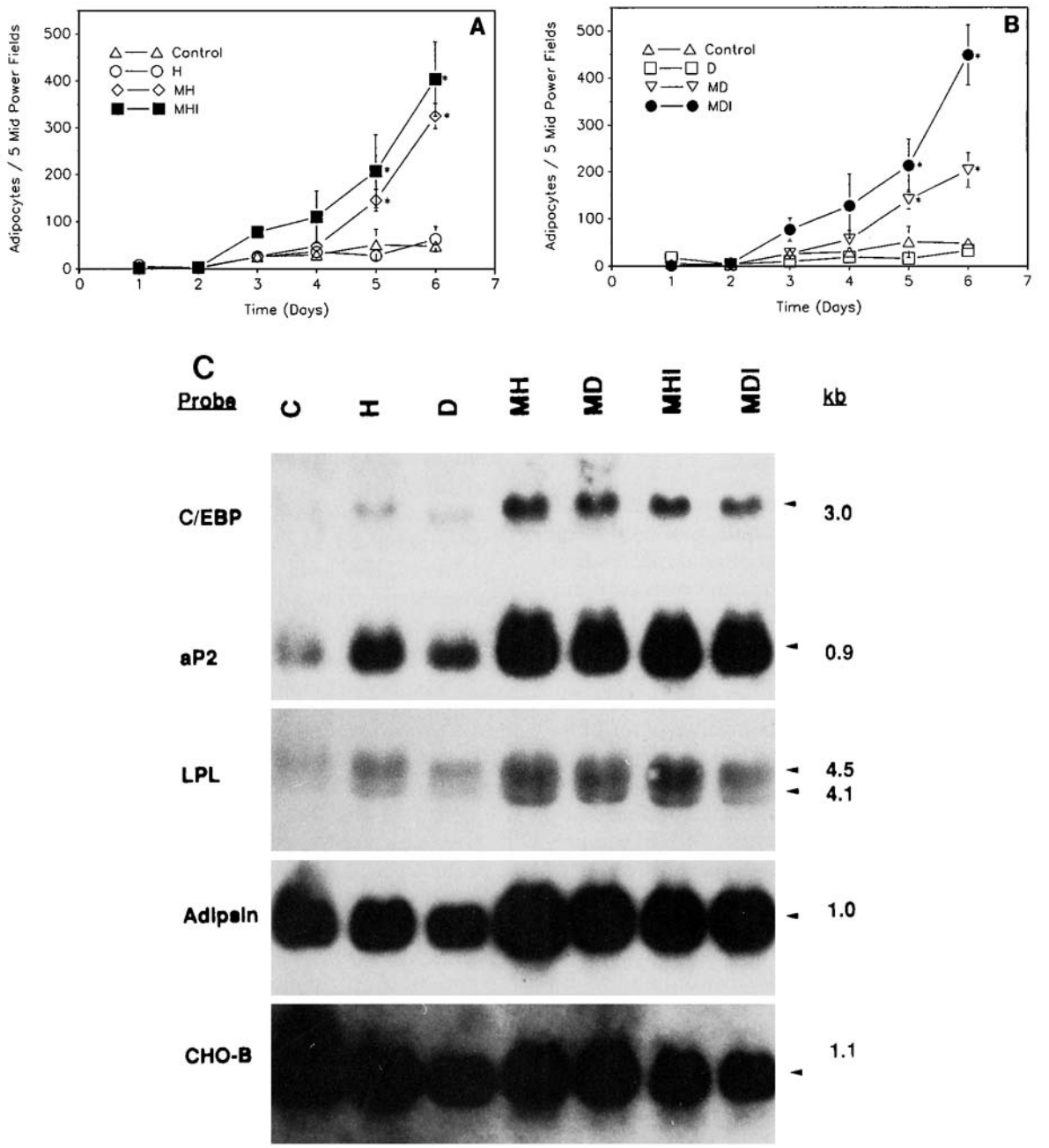



**Fig. 3.** Response of +/+2.4 cells to adipogenic agonists. Confluent and quiescent cultures of +/+2.4 cells were treated for a period of 60 h (days 0–2) with varying combinations of adipogenic agonists. The following concentrations were employed: hydrocortisone (H), $5 \times 10^{-7}$ M; dexamethasone (D), $5 \times 10^{-7}$ M; MIBX (M), 0.5 mM; indomethacin (I), 0.06 mM. Whenever MIBX was present, insulin was added at a concentration of 5 mcg/ml as described in Materials and Methods. Cells were treated with the following combinations and are represented by the indicated symbol: untreated controls (C), open triangle; hydrocortisone alone (H), open circle; dexamethasone alone (D), open square; MIBX + hydrocortisone (MH), open diamond; MIBX + dexamethasone (MD), inverted triangle; MIBX + hydrocortisone + indomethacin (MHI), closed square; MIBX + dexamethasone + indomethacin (MDI), closed circle. The data in A and B represent the mean ± S.D. of 4 separate individual data points and are representative of 2 separate experiments. Asterisks indicate $P < 0.05$ relative to the control based on the student t test. **A:** Induction of adipocyte differentiation in the presence of hydrocortisone. The number of phenotypic adipocytes was determined in 5 separate phase contrast mid-power (×20) fields using a Zeiss IM 35 microscope. **B:** Induction of adipocyte differentiation in the presence of dexamethasone. **C:** Northern blot analysis of total RNA from induced cells. The individual cultures examined in A and B were harvested for total RNA 6 days after induction. Northern blots were hybridized successively with probes for the adipocyte specific genes C/EBP, aP2, LPL, and adipsin; the constitutively expressed gene CHO-B was utilized as a control for relatively equal loading between lanes. Transcript sizes are indicated (kb). Representative of 4 separate studies.

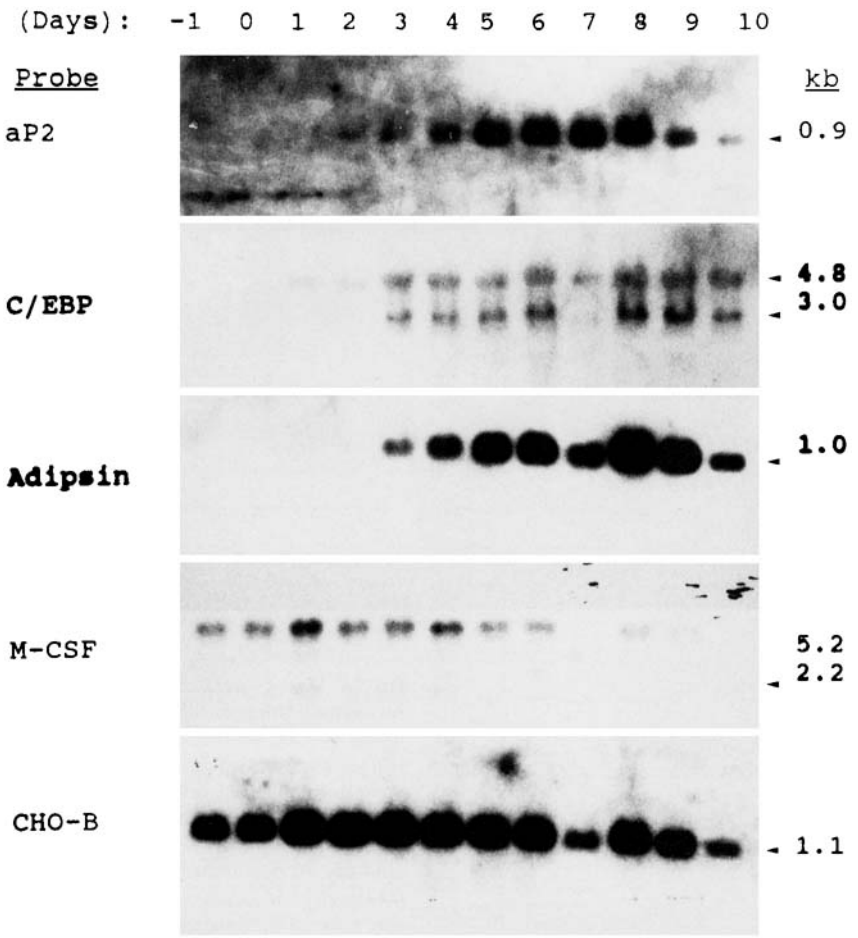
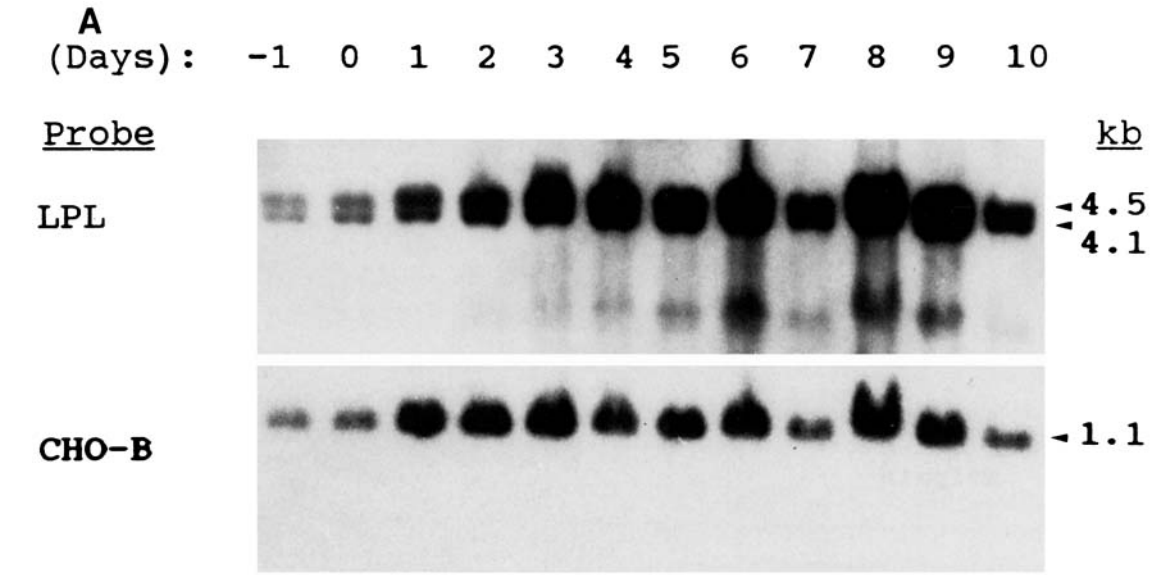



**Fig. 4.** Kinetics of adipocyte marker gene steady state mRNA levels. Total RNA was harvested from +/+2.4 cells from 1 day before (−1) until 10 days (10) after induction with methylisobutylxanthine/hydrocortisone/indomethacin/insulin. Approximately 10 mcg total RNA/lane was examined on Northern blots successively hybridized with cDNA probes for aP2, adipsin, C/EBP, M-CSF, and CHO-B as a control. The transcript sizes (kb) are indicated. Data is representative of 2 separate experiments.

## DISCUSSION

The +/+2.4 stromal cell clone undergoes spontaneous adipogenesis which was accelerated following treatment with glucocorticoids, MIBX, and indomethacin. The +/+2.4 cell line was originally defined based on its ability to support myelopoiesis and this function was retained in the current passage. However, while these cells produced levels of IL-7 mRNA detectable by PCR, they did not support the growth of IL-7 dependent B lineage lymphoid cells. Adipogenesis was accompanied by increased steady state mRNA levels for adipocyte specific gene markers. For the genes aP2, adipsin, and C/EBP, expression was not detected in cultures lacking morphologically identifiable adipocytes. Following induction of adipogenesis, the maximum signal intensity for these transcripts was detected after 6–8 days. Although the pre-adipocytes constitutively expressed some LPL mRNA, both the LPL mRNA steady state level and enzyme activity were elevated in a time-dependent manner following adipocyte induction. In contrast, the M-CSF steady state mRNA levels decreased slightly following induction of adipogenesis. This is consistent with observations in other stromal cell lines [Watanabe and Takano, 1991].

Bone marrow adipocytes are thought to differ from extramedullary or white adipose tissue (WAT) based on the response of the bone marrow to lipolytic activation. Studies of long-term bone marrow cultures in vitro indicated that adipocytes do not respond to insulin [Greenberger 1978, 1979]. In whole animal studies, the



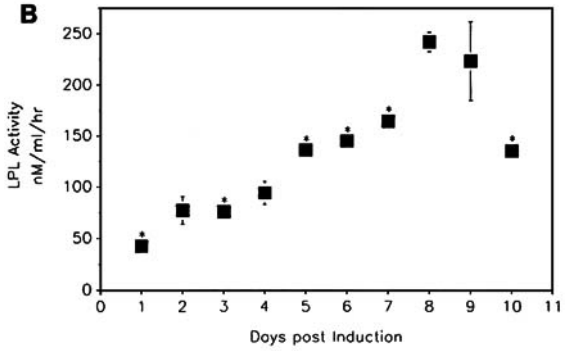




**Fig. 5.** Kinetics of LPL expression. **A:** Northern blot analysis. An identical Northern blot to that described in the legend of Fig. 4 was probed successively with a murine cDNA LPL probe (cloned as described in Materials and Methods) and the CHO-B control. Transcript sizes (kb) are indicated. **B:** Lipoprotein lipase enzyme activity. The heparin releasable LPL enzyme activity was determined in the supernatant of +/+2.4 cells from day 1 until day 10 post-induction. Values represent the mean +/− the standard deviation of 2 separate experiments, each of which was assayed in triplicate. Asterisks indicate values where the standard deviation error bars were less than 10 nM/ml/h.

bone marrow adipose stores are not depleted by acute starvation, in comparison to renal fat pads [Bathija et al., 1979]. Long-term starvation eventually causes bone marrow fat atrophy and is accompanied by deposition of extracellular mucopolysaccharides [Mant and Faragher, 1972; Seaman et al., 1978; Tavassoli et al., 1976]. However, in experimentally induced anemia, erythropoiesis correlates with a dramatic decrease in bone marrow adipose stores, presumably through the release of locally acting lipolytic agents [Bathija, 1979; Maniatis et al., 1971]. In contrast, suppression of erythropoiesis by hypertransfusion results in a higher percentage of adipose tissue within the bone marrow cavity [Brookoff and Weiss, 1982]. Despite these differences between bone marrow and white adipose tissues, the bone marrow does play a role in energy metabolism. In chylomicron clearance studies, the bone marrow accounted for a significant proportion of chylomicron uptake in all species examined [Hussain et al., 1989a, 1989b]. Likewise, the turnover rate for free fatty acids was approximately five times greater in bone marrow compared to perinephric adipose tissue, consistent with a greater rate of lipolysis [Trubowitz and Bathija, 1977].

A number of murine cell lines (3T3-L1, TA-1, Ob17) have provided in vitro models for adipogenesis and its regulation [Ailhaud, 1982; Beutler et al., 1985; Chapman et al., 1984; Cornelius et al., 1988; Dani et al., 1990; Green and Kehinde, 1975, 1976; Ogawa et al., 1988; Price et al., 1986a,b; Torti et al., 1985, 1989; Zechner et al., 1988]. These lines have been derived either directly from adipose tissue or from fetal fibroblasts [Ailhaud, 1982]. Although many bone marrow pre-adipocyte stromal cell lines have been cloned, studies examining their adipocyte differentiation at the molecular level are limited [Gimble et al., 1989, 1990; Gimble, 1990; Grigoriadis et al., 1990; Watanabe and Takano, 1991]. In the lymphopoietic-supporting BMS2 stromal cell line, adipogenesis correlated with the induction

of aP2, adipsin, and LPL steady state mRNA levels as well as increased enzyme activity for glycerophosphate dehydrogenase (GPD) and LPL [Gimble et al., 1990]. The presence of antagonists such as TNF and TGF beta blocked BMS2 differentiation [Gimble et al., 1989]. In the myelopoietic-supporting H-1/A stromal cell line, M-CSF steady state mRNA levels decreased following adipocyte differentiation due to post-transcriptional regulation [Watanabe and Takano, 1991]. Adipogenesis in the +/+2.4 cells resembles these other murine stromal cell lines with respect to inducing agents and gene induction. Likewise, the kinetics of adipocyte marker gene expression parallel those reported in pre-adipocyte cell lines of extramedullary origin [Chapman et al., 1984; Christy et al., 1989; Spiegelman et al., 1983]. Together, these results demonstrate that the mechanism of adipogenesis in myelopoietic-supporting bone marrow stromal cells has features in common with lymphopoietic-supporting stromal cells and WAT cell lines.

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