Analysis of saturated fatty acid (FA) in cultured fibroblasts is used for the diagnosis of X-linked Adrenoleukodystrophy (X-ALD). The culture mediums normally used are basic mediums such as Hank's, Eagle's or RPMI plus calf serum. Due to a variable expression of ALDP (the defective protein in X-ALD) observed in cells cultured in different mediums (Ruiz et al. 1997), we have studied the influence of the culture medium in the biochemical parameters used in X-ALD diagnosis. We have focused our attention in FA profile.

1. CULTURE MEDIUMS

The mediums studied were: a) Chang C medium (Irvine, Inc) and Amniomax (Life Technologies Inc.) used to enhance growth for karyotypic studying in prenatal testing and b) minimum essential medium Eagle (Sigma Chemical Co) supplemented with 15% foetal bovine serum (Gibco BRL) (MEM+FB), used for a wide variety of cell grown in monolayers.

FA analysis was made by direct transesterification method described by Lepage and Roy (1986). Separation and identification was performed by gas-liquid chromatography.

In the analysis of the mediums we found greater FA concentration in Chang than in MEM+FB or Amniomax (x2). FA series distribution was, in Chang: \( \omega 6 > \text{saturated} > \omega 9 > \omega 3 \) and in MEM+FB or Amniomax: saturated > \( \omega 9 > \omega 6 > \omega 3 \) (Fig. 1). Chang and Amniomax remained unchanging within batches, but MEM+FB showed some variability in \( \omega 6 \) and \( \omega 9 \) series percentage. Similar FA composition in each series was found, except for \( \omega 6 \);
the 18:2/20:4ω6 ratio was 10:1 in Chang and 1:1 in MEM+FB and Amniomax (Fig. 2).

![Graph showing FA series distribution in different culture mediums](image)

*Figure 1. FA series distribution (%) in different culture mediums*

![Graph showing FA composition of ω6 series in different mediums](image)

*Figure 2. FA composition of ω6 series in different mediums. FA are expressed as % in the series and total ω6 series concentration as μg/ml*

2. **FIBROBLASTS**

The FA in control fibroblasts (n=3) cultured in MEM+FB, Amniomax and Chang, showed identical total FA concentration, and the same 18:2/20:4ω6 ratio (1:3). The fact that 18:2/20:4ω6 ratio remained unchanging in cells in spite of the differences in the mediums suggests activation of proteins, as desaturases and elongases, involved in the homeostasis of FA synthetic pathway. Regarding FA series distribution, we observed different behaviour depending on the medium: in cells cultured in