

REPORT 30

Light and electron microscopy findings in subcutaneous fat in antiretroviral treated and HIV-infected patients

This is a small study looking at biopsies of fat cells from 3 NRTI+PI treated individuals and 1 NRTI-treated but PI-naïve individual with fat loss and accumulation. The biopsies were examined by electron and light microscopy. The authors found abnormalities which are described below. It's important to bear in mind that this is a very small study limiting conclusions. The author concludes that NRTI treatment alone can lead to the abnormalities found, and the addition of PI treatment may increase abnormalities.

A number of studies have been reported in which people with NRTI experience but without PI experience have experienced body changes (lipodystrophy). There may be a number of potential contributing causes leading to a person having these body changes which we generally refer to as lipodystrophy including genetic predisposition, diabetes, insulin resistance, HIV viral load, change in HIV viral load due to therapy, age, diet, and the effect of HIV on a person's immune system. A number of studies have suggested that d4T may be more of a culprit than other NRTIs, except for ddC but ddC is not really used much anymore. However, some studies were unable to discern a difference between d4T and other NRTIs in causing lipodystrophy. A number of researchers think the studies pointing to d4T are confounding and not conclusive. For example, often these studies don't appear to be able to account for AZT experience prior to starting AZT. Some researchers think there may be two distinct syndromes--one leading to fat wasting and the other leading to fat accumulation. A number of researchers think NRTIs may be the cause of lipodystrophy and protease inhibitors may lead to fat accumulation, but these are hypotheses. Much research effort is being focused on body changes and I think slow progress is being made, but we do not have answers yet to some of these burning questions: what causes body changes and how can we prevent them or intervene to reverse it?

At the 7th European Conference on Clinical Aspects and treatment of HIV-infection in Lisbon in October '99, the same author of this paper (Mallal) reported on a study from which he concluded chronic NRTI use appears to predispose the patient to fat loss, which is accelerated when protease inhibitors and NRTIs are combined. In the study reported on at Durban he used the microscopy testing and reached the same conclusions.

In this study reported at Durban by Mallal he doesn't report any other information about the patients except some of their treatment experience. But he doesn't discuss their history of viral load, genetics, insulin resistance, etc. This makes interpretation more complicated.

Simon Mallal and D Nolan from the Royal Perth Hospital in Perth Australia, authors of this late breaker poster at Durban, wrote that histological features of subcutaneous fat in PI-treated patients affected by the lipodystrophy syndrome have been recently described. Here we compare the ultrastructural features of affected fat in PI-treated and PI-naïve patients with clinical evidence of fat loss as well as accumulation. Excision biopsies were taken from the supra-iliac region of 4 affected patients including one PI-naïve person. One specimen was also taken from a buffalo hump. Samples were also taken from 3 untreated HIV+ persons for comparison.

Using light microscopy in PI-treated and PI-naïve, mature adipocytes were present, with moderate numbers of lipogranulomata seen in affected patients, associated with adipose (fat cells) cell loss and increased variation in adipocyte size. Apparent increased vasculature without evidence of vessel proliferation. Samples from PI-naïve patient and PI-treated patients showed identical changes with no quantitative differences in lipogranulomata between the two groups, and identical features also seen in buffalo hump. There was no evidence of immature adipocyte forms, or of brown fat. Lipid-laden macrophages present in lipogranulomata in the absence of other inflammatory cells.

In HIV-infected but untreated patients, subcutaneous biopsy from one patient showed occasional lipogranulomata, with normal adipose light microscopy morphology in remaining two patients.

Using electron microscopy in PI-treated patients they found expanded adipocyte cytoplasm containing multiple lipid droplets;

mitochondria abundant, with abnormal budding and elongated forms; mitochondrial cristae consistently abnormal with whorled forms and disoriented axis, although with no evidence of paracrystalline inclusions. Redundant basal lamina of adipocytes and blood vessels frequently noted, consistent with cellular atrophy. Nuclear chromatin pattern normal, with no changes to nuclear envelope observed. Deposition of electron-dense granular material was noted in the periphery of adipocytes, located with the cell membrane. This correlated with brown pigment of fat noted at operation, and brown pigment seen at the periphery of some adipocytes on light microscopy. This was submitted for energy dispersive X-ray spectroscopy, which indicated that granules are organic rather than metallic. Negative PAS staining has excluded glycogen deposition. One possibility is that this material may be non-esterified (free) fatty acid desposition

In PI-naïve patients adipocytes showed less intense expansion of cytoplasm and proliferation of mitochondria, although elongated and budding mitochondria were noted. No abnormalities of cristall orientation were observed. Intracytoplasmic lipid droplets were present. Redundant basal lamina noted on blood vessels consistent atrophic change, although this did not affect adipocytes. No peripheral granular deposition noted.

HIV-infected untreated patients: no ultrastructural abnormalities noted, despite presence of occasional lipogranulomata on light microscopy

Mallal concluded that NRTI therapy with or without PI therapy is associated with adipocyte (fat cells) cell loss and lipogranulomata at the light microscopy level, with identical morphologic features seen in regions of fat loss and accumulation. While untreated HIV-infected patients had normal adipocyte ultrastructure, patients affected by subcutaneous fat wasting consistently demonstrated abnormal mature adipocytes with prominent mitochondrial abnormalities, cytoplasmic lipid accumulation, and adipocyte atrophy. NRTI therapy appears to be a sufficient cause of the primary fat pathology, although in this limited study ultrastructural changes appeared to be more severe in the presence of PI therapy.