Hyaluronic acid is an unbranched high molecular weight polysaccharide, consisting of repeating disaccharide units of glucuronic acid - 1, 3N acetyl glucosamine linked by 1-4 bonds.

The introduction of sensitive assays has now made it possible to detect HA in very low concentrations (Engström-Laurent et al., 1985a; Delpech et al., 1985). Tissue HA enters the circulation via the lymph and is rapidly extracted and catabolised by the liver (Laurent & Fraser, 1986), resulting in a normal serum HA level of 10–100 μg l⁻¹. The level of HA can rise in cirrhosis (Engström-Laurent et al., 1985b) and rheumatoid arthritis (Engström-Laurent & Hällgren, 1985) and in end stage renal failure (Hällgren et al., 1987).

There have been a few reports of raised serum levels of HA in advanced cancer; Delpech et al. (1985) made this observation when developing their enzyme immunoassay. Later they used this test to investigate the HA concentrations in the serum and pleural effusions in mesothelioma and observed high levels in advanced disease (mean 750 μg l⁻¹; range 29–5,833 μg l⁻¹), but the test was unsuitable for early diagnosis (Fremourg et al., 1987). Manley & Warren (1987) using a nephelometric assay also observed raised serum HA levels in cancer, but their technique gave much higher concentrations for normal HA levels (mean 1.09 mg l⁻¹; range 0–4 mg l⁻¹).

The availability of a commercial radiometric (HA test 50, Pharmacia, Uppsala, Sweden) has provided an opportunity to extend these preliminary observations. This assay has been developed from the method described by Tengblad (1980) and Lauret & Tengblad (1980).

The test is based on the use of specific hyaluronic acid binding proteins, (HABP) isolated from bovine cartilage. First the hyaluronic acid from the sample is allowed to bind ¹²⁵I-labelled HABP in solution for at least 60 min. The unbound ¹²⁵I-HABP is then quantitated by incubating with HA covalently coupled to sepharose particles of small size and low density. The particles stay suspended in the 45 min incubation. Separation is performed by centrifugation followed by decanting. The radioactivity bound to the particles is measured. It is inversely proportional to the concentration of HA in the sample. The assay has a detection limit of <5 μg l⁻¹, and an operating range of <5–500 μg l⁻¹. The coefficient of variation within and between assays was 6.5 and 4.9% respectively for a sample with a mean value of 29.0 μg l⁻¹, 5.2 and 8.0% respectively for a sample with a mean value of 312 μg l⁻¹. The assay measures HA with a wide range of relative molecular weights ranging from <10⁴–5 x 10⁵ KD. The manufacturer’s data indicate that there is a marked age dependency for the normal values rising from a mean of 18–69 μg l⁻¹ as age rose from <20–>60 years old. We chose our controls from healthy persons between 42 and 64 years of age.

The distribution of serum HA values in the controls and 121 adult patients with metastatic cancer or large local tumours at presentation is shown in Figure 1. It is evident that within each type of cancer individual patients can show strongly elevated levels of HA, but statistical significance (Spearman rank test) for an overall increased level compared to the controls was only present in pancreatic cancer (P = 0.0097), small cell lung cancer (P = 0.01) and carcinoma of the prostate (P = 0.0069). Sera from 20 patients with hepatic metastases gave a median HA of 74 μg l⁻¹ and range, 21–970 μg l⁻¹; and sera from 20 patients with metastases involving bone had a median HA of 66.5 μg l⁻¹ and range, 3–789 μg l⁻¹; both were not significantly different to normal by a non-parametric analysis.

Whilst the patients with raised HA (>130 ng l⁻¹) tended to have other tumour markers that were raised, there were other patients with normal HA levels and raised marker levels, e.g., in carcinoma of the pancreas the HA vs. CEA (>5 ng ml⁻¹) 9:12, in colorectal 2:6, in prostate cancer vs. PSA (>10 ng ml⁻¹) 7:11 in small cell lung cancer vs. NSE (>13 ng ml⁻¹) 8:18. The induction of clinical and biochemical remission in metastatic prostatic cancer by hormone manipulation was not associated with any significant change in HA level as shown by a paired t-test for 9 paired values (P = 0.2865).

The levels of serum HA are age dependent; in persons <20 years the 95th percentile is 37 μg l⁻¹. When sera from patients with untreated Wilms’ tumour or neuroblastoma were measured it was evident that Wilms’ tumour patients had a high incidence of greatly raised HA levels; in over a third the levels were >500 μg l⁻¹, and in 4 of them >20,000 μg l⁻¹. In neuroblastoma, by contrast, the pattern was similar to that seen in adult cancers with a median level of 25 μg l⁻¹ and range, 11–327 μg l⁻¹ (Table I). The raised levels of HA in Wilms’ tumour did not appear to be stage dependent, levels >500 μg l⁻¹ occurring in some patients with all stages. The molecular weight distribution of HA in the sera of 2 patients with Wilms’ tumours was analysed by high performance gel chromatography (Superose 6, Pharmacia). Both samples showed that the predominant form of HA was a high molecular weight polymer (>10⁷ KD) and this was associated with a wide range of smaller polymers to ~10⁴ KD (Figure 2).

These studies confirm that a raised serum HA may accompany malignant disease but cannot be considered as a

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**SHORT COMMUNICATION**

**Serum hyaluronic acid levels in cancer**

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Research

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