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Use of biomarkers of occupational musculoskeletal disorders in epidemiology and laboratory animal model development

by J Patrick Mastin, PhD,[1] Gerry M Henningsen, DVM,[1] Lawrence J Fine, MD[2]

Advances in biotechnology and in the knowledge of molecular and cellular functions related to physiological and pathological changes in musculoskeletal tissues are now providing the opportunity to relate biochemical changes with the onset and progression of musculoskeletal diseases. Modern methods, used in conjunction with classical techniques, to evaluate these potential biomarkers include immunochemistry, automated high-resolution chromatography, electrophoresis, and other bioanalytical procedures enhanced by the use of advanced optics, lasers, and computers. Sufficiently defined biomarkers have the potential to detect subclinical musculoskeletal disease and monitor the severity of musculoskeletal disease in individuals and populations.

Epidemiologic studies have identified associations between certain occupations and musculoskeletal disorders, including disorders of the lower extremities, back, upper extremities, and neck, that can involve nerve compression, inflammation of specific tendons or muscles, cartilage degeneration, or unknown or uncertain mechanisms. Many of these musculoskeletal diseases are chronic, progressive disorders that can lead to impairment and disability and possibly to irreversible damage. Biomarker technology offers the possibility of detecting musculoskeletal disease at an early, preclinical stage and allowing intervention measures to be taken before irreversible damage occurs. To date, however, little work has been done to identify and use biomarkers for the diagnosis of early musculoskeletal disorders.

A major obstacle to finding biomarkers of musculoskeletal disorders in the general population is that the subjects are typically drawn from relatively heterogeneous populations in regard to risk factors for musculoskeletal disease. When one deals specifically with occupational musculoskeletal disease, however, the subjects are usually from a more homogeneous population comprised of reasonably healthy individuals of work age who have a known risk for musculoskeletal disorders due to their occupations. Conducting research on worker populations at risk of musculoskeletal disorders should facilitate the identification of biomarkers of musculoskeletal disease.

Types of biomarkers of musculoskeletal disease

In general, there are four types of biomolecules that have been investigated as potential biomarkers of musculoskeletal disease, namely, (i) proteins that are constituents of musculoskeletal tissue and are either released through catabolic processes or produced as part of the repair process (eg, cartilage proteins from joints), (ii) proteins associated with the inflammatory response (eg, acute phase proteins), (iii) preinflammatory response biomolecules (occurring before the inflammatory response and often inducing it), and (iv) indicators of immune-mediated (autoimmunity or hypersensitivity) inflammatory processes. It should be realized that most musculoskeletal biomarkers are not specific for a particular disorder, but they can be compatible with and aid in supporting the presence and extent of disease when used as part of a profile of appropriate tests and accurate work histories. Occasionally, more specific biomarkers can be found for certain musculoskeletal disorders, such as for autoimmune disorders. Of the musculoskeletal disorders with possible occupational etiology, only potential biomarkers of osteoarthritis have been extensively studied. Since certain other occupational musculoskeletal disorders are also chronic and progressive in nature, biomarkers for these conditions might well be identified in the future.

Most investigations of potential biomarkers of osteoarthritis have focused on the presence of cartilage components in serum, primarily proteoglycans and glycosaminoglycans (especially keratan sulfate). Fife & Brandt (1) and Fife (2) have also identified a noncollagenous glycoprotein (cartilage matrix glycoprotein) in articular cartilage that appears to be released from degenerating cartilage. Other researchers have found changes in inflammatory proteins (eg, acute phase proteins) in serum from patients with osteo-

[1] Applied Biology Branch, Division of Biomedical and Behavioral Science, National Institute for Occupational Safety and Health Cincinnati, Ohio, United States.

[2] Division of Surveillance, Hazard Evaluations, and Field Studies, National Institute for Occupational Safety and Health Cincinnati, Ohio, United States.

Correspondence to: Dr JP Mastin, National Institute for Occupational Safety and Health, 4676 Columbia Parkway, Cincinnati, OH 45226, USA.

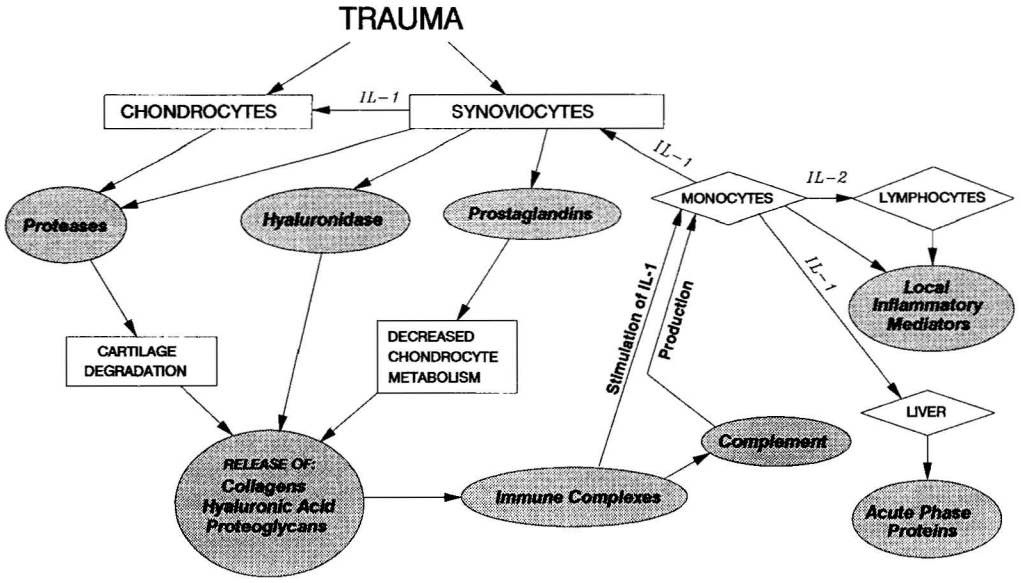

Figure 1. Paradigm for the development of degenerative joint disease resulting from trauma (adapted from references 11 and 12). (ellipses = potential biomarkers, IL-1 = interleukin 1, IL-2 = interleukin 2)

arthritis (3, 4). Finally, researchers have investigated antibodies and cellular immune reactions directed against components of cartilage (eg, collagen, link protein, and chondrocyte membrane antigens) as possible biomarkers of osteoarthritis (5—8). Although low-back pain is one of the most common musculoskeletal disorders in this country, there has been very little research on biomarkers of early back disease. Decreased plasma and saliva levels of substance P, a neurochemical having inflammatory properties, have been reported for patients with chronic back pain (9). Therefore, substance P might prove to be a useful biomarker of back pain with certain etiologies. It is also likely that components of intervertebral disks are released into systemic circulation, along with the resultant mediators of inflammation, as a result of chronic back injury and, hence, could serve as biomarkers of this condition.

Cumulative trauma disorders are chronic, sometimes progressive, disorders, and intervention procedures could be very effective if the conditions could be detected early (eg, through the detection of early biomarkers). To our knowledge, no specific research in the area of biomarkers of cumulative trauma disorders has been published. Amyloid has been detected in the tenosynovium of the wrist in some patients with carpal tunnel syndrome (10). Amyloid protein or precursor proteins may serve as a starting point for research to identify biomarkers of cumulative trauma disorders with certain etiologies. In addition, since cumulative trauma disorders are local inflammatory disorders, it is likely that changes in serum inflammatory proteins (eg, acute phase proteins) may be useful as biomarkers of certain types of cumulative trauma disorders.

Future research needs

Several lines of research should be included in future developmental studies of biomarkers of occupational musculoskeletal disease. First, efforts should proceed to apply emerging technologies to develop more sensitive and specific assays for potential biomarkers. Second, efforts to understand the pathophysiology of occupational musculoskeletal disorders should continue, especially for chronic, progressive ones (eg, cumulative trauma disorders), and especially in the early stages. Figure 1 shows one plausible paradigm for the development of degenerative joint disease as a result of trauma. Investigators have identified the biochemicals enclosed in the ellipses as ones that are produced as a result of degradative or inflammatory processes. These biomolecules could be investigated as possible biomarkers of trauma-induced degenerative joint disease. Finally, the development of animal models to be used in biomarker research on musculoskeletal disease and the validation of these markers in selected human populations should continue.

Current research

An example of this research approach is currently underway at the National Institute for Occupational Safety and Health. The purpose of the project is to identify biomarkers of the early stages of experimental progressive joint disease in two animal models. In one

model, a predominantly inflammatory lesion is induced in rabbits through the immobilization of one leg in extension for one, four, or seven weeks. Blood samples are collected before the splints are applied, at weekly intervals, and at necropsy for analysis with gel electrophoresis, liquid chromatography, and immunoassays to identify potential biomarkers. In the second model, a predominantly degenerative lesion is induced in juvenile rabbits by the daily oral administration of nalidixic acid for 3, 7, or 14 d. Blood samples are collected prior to the initial dosing and at sacrifice, and they are analyzed as in the first model. These two models were chosen because they produce joint lesions that are similar to the pathological processes seen in humans — inflammation and degeneration.

Preliminary results of this research identified four potential biomarkers for immobilization-induced joint disease: haptoglobin, beta$_2$-microglobulin, cartilage matrix glycoprotein, and a 25-kilodalton glycoprotein (perhaps related to haptoglobin). Details of these studies will be presented later. Efforts are currently being directed towards the further investigation of these and other potential biomarkers and towards the development of sensitive, relatively simple assays (enzyme-linked immunosorbent assay, etc) for the routine measurement of these biomolecules in both human and animal serum samples. A field study of workers at risk for occupational joint disease (eg, carpet layers) will be conducted to validate, in a human population, the potential biomarkers of degenerative joint disease identified in these animal studies. Ultimately, the aim is to identify and characterize biomarkers of work-related joint diseases at the earliest, preclinical and reversible stages to permit successful intervention or preventive measures.

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