

# Macrophagic Myofasciitis in Children Is a Localized Reaction to Vaccination

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Macrophagic myofasciitis is a novel, "inflammatory myopathy" described after a variety of vaccinations, almost exclusively in adults. We examined the relevance of histological findings of this myopathy to the clinical presentation in pediatric patients. Muscle biopsies from 8 children (7 months to 6 years old) with histological features of macrophagic myofasciitis were reviewed and correlated with the clinical manifestations. Patients underwent quadriceps muscle biopsy for suspected mitochondrial disease (4 patients), spinal muscular atrophy (2 patients), myoglobinuria (1 patient), and hypotonia with motor delay (1 patient). All biopsies showed identical granulomas composed of periodic acid-Schiff–positive and CD68-positive macrophages. Characteristic aluminum hydroxide crystals were identified by electron microscopy in 2 cases. The biopsy established diagnoses other than macrophagic myofasciitis in 5 patients: spinal muscular atrophy (2), Duchenne muscular dystrophy

(1), phospho-glycerate kinase deficiency (1), and cytochrome *c* oxidase deficiency (1). Three children with manifestations and/or a family history of mitochondrial disease had otherwise morphologically normal muscle. All children had routine vaccinations between 2 months and 1 year before the biopsy, with up to 11 intramuscular injections, including the biopsy sites. There was no correlation between histological findings of macrophagic myofasciitis in biopsies and the clinical symptoms. We believe that macrophagic myofasciitis represents a localized histological hallmark of previous immunization with the aluminum hydroxide adjuvants contained in vaccines, rather than a primary or distinct inflammatory muscle disease.

**Keywords:** macrophagic myofasciitis; inflammatory myopathy; vaccination; aluminum

Macrophagic myofasciitis was initially reported in adult patients with myalgia, arthralgia, weakness, and fever, and histologically documented collections of periodic acid-Schiff–positive macrophages accompanied by lympho-plasmocytic infiltrates in muscle biopsies.<sup>1-4</sup> Electron microscopy of these macrophages demonstrated a characteristic intracytoplasmic inclusion identified by mass spectrometry as aluminum hydroxide, an adjuvant used in vaccines.<sup>1-7</sup> Because the patients were vaccinated 3 to 96 months before biopsy, the development of the muscle lesions and the clinical symptoms were attributed to prolonged tissue retention and an abnormal immunological response to aluminum hydroxide.<sup>1-4</sup> The largest published series of patients originated from France after an anti-hepatitis B vaccination campaign.<sup>1-4</sup>

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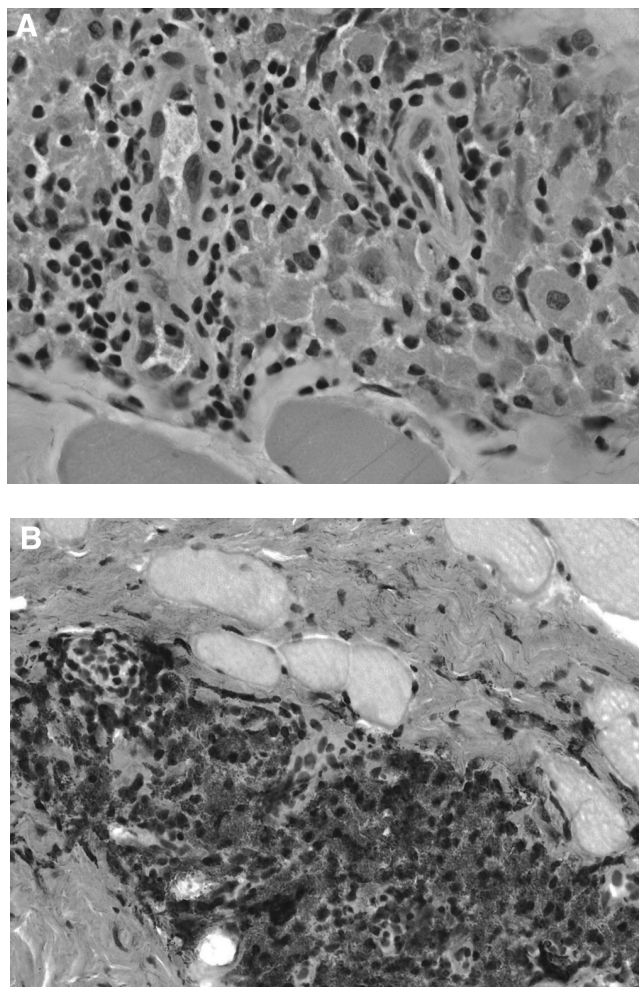
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Subsequently several cases of this disease were reported from other countries.<sup>5-15</sup> Despite universal vaccination programs, myofasciitis has been rarely observed in children.<sup>6-12,14,15</sup> Histological changes of myofasciitis were considered as significant in some of the pediatric cases<sup>10,12</sup> or regarded as an incidental morphological finding in other series of patients.<sup>6-8,11,14</sup> We report the clinical and pathological findings in 8 children with biopsy-proven macrophagic myofasciitis, overlapping with a variety of neuromuscular disorders.

## Patients and Methods

All patients with characteristic pathological changes in muscle biopsy (Figures 1 and 2) were retrospectively studied. After obtaining informed consent, these patients had undergone open quadriceps femoris biopsy under local anesthesia to evaluate suspected neuromuscular disorders. Clinical and pathological diagnoses as well as basic patient information are summarized in Table 1. The biopsy specimens were snap frozen in isopentane cooled with liquid nitrogen at  $-70^{\circ}\text{C}$ . Enzyme histochemistry (ATPase [at 9.4, 4.6, and 4.3], reduced nicotinamide adenine dinucleotide (NADH), succinic dehydrogenase, acid and



**Figure 1.** Case 7. Phospho-glycerate kinase deficiency. A, Hematoxylin-eosin–stained section showing epithelioid histiocytic cells with minimal lymphocytic component. B, Deeper level of the same lesions display granular cytoplasmic immunoreactivity for chymotrypsin.

alkaline phosphatase, and nonspecific esterase) and routine staining (hematoxylin–eosin [H&E], modified Gomori trichrome, and periodic acid–Schiff reaction) were performed using established techniques. Immunohistochemistry for the following antigens was carried out: CD68, CD20, CD3, CD8, S-100 protein, trypsin–antitrypsin, antichymotrypsin, factor XIIIa, MIB-1, and CD1A, using a Ventana immunostainer and supplied antibody kits and/or Novocastra primary antibodies in the recommended dilutions. In cases 3 and 5, with severe myopathic changes, supplementary immunohistochemistry was performed for the following components: dystrophin (central, carboxyl, and amino domains);  $\alpha$ -,  $\beta$ -,  $\gamma$ -, and  $\delta$ -sarcoglycan;  $\beta$ -dystroglycan; merosin; and spectrin; each with positive and negative controls. Small tissue pieces were fixed in 1.6% glutaraldehyde for electron microscopy, and additional samples were snap frozen for biochemical studies (subsequently carried out



**Figure 2.** Case 7. Characteristic spicular inclusions of aluminum hydroxide within a macrophage (original magnification  $\times 39\,000$ ).

only in case 7). In case 8, only paraffin-embedded material was available, limiting examination to inflammatory cell typing.

## Results

All the biopsies displayed characteristic focal collections of periodic acid-Schiff–positive and oil red O–positive epithelioid macrophages, slightly basophilic cytoplasm reactive for CD68, trypsin-antitrypsin, antichymotrypsin (Figure 1), acid phosphatase, and nonspecific esterase. In 3 biopsies (cases 2, 7, and 9), these changes were accompanied by rare B-cell and T-cell infiltrations, representing less than 10% of the inflammatory cell population. Biopsies from cases 1 and 3 also displayed few, small nodular collections of CD-45–positive cells composed of a mixture of CD3-, CD8-, CD20-, and CD68-immunoreactive cells. In case 8, macrophagic infiltrate was limited to the perimysium and fascia. Myonecrosis in conjunction with granulomas was present in 2 cases (4 and 5). Occasional regenerating fibers were observed in all biopsies. In 1 case, macrophage infiltrations were accompanied by intense fibrosis (case 8).

In addition to collections of macrophages, the biopsies in case 1 and 3 showed groups of highly atrophic rounded fibers, fiber type grouping, and markedly enlarged type I fibers, consistent with spinal muscular atrophy, subsequently confirmed by genetic testing. Cases 2, 4, and 6, with the clinical diagnosis of mitochondrial diseases, showed only minimal variability in size and shape of individual muscle

**Table 1.** Summary of Clinical Information and Pathological Findings<sup>a</sup>

Case	Age/Sex	Clinical Findings and Suspected Diagnosis	Additional Muscle Biopsy Findings <sup>a</sup>	EM	Immunization <sup>b</sup>	Consanguinity
1	7 mo/M	Spinal muscular atrophy with respiratory failure, on ventilator	Fiber type grouping, large round fibers. Spinal muscular atrophy	Granuloma not in sample	DPT Hepatitis B (6 mo)	–
2	9 mo/M	Mitochondrial disease. Microcephaly. Delayed milestones. Family history of mitochondrial disease	Nonspecific minimal pathological changes	Granuloma not in sample	DPT, Hepatitis B (6 mo)	+
3	18 mo/M	Hypotonia. Delayed milestones. Spinal muscular atrophy	Type grouping, large round fibers. Spinal muscular atrophy	Granuloma not in sample	DPT MMR Hepatitis B H. influenza Meningitis (15 mo)	–
4	2 y 3 mo/F	Spasticity. Lactic acidosis from age of 9 months. Family history of lactic acidosis	Focal necrosis around granuloma. Normal muscle	NA	DPT, MMR Hepatitis B H. Influenza (18 mo)	+
5	3 y/M	Progressive weakness and elevated muscle enzymes	Dystrophin negative Duchenne muscular dystrophy	Granuloma not in sample	DPT, MMR Hepatitis B H. Influenza (18 mo)	–
6	2 y/F	Dysmorphic face. Lactic acidosis. Psychomotor retardation. Microcephaly. Family history of neurological disease and lactic acidosis	COX deficiency. Denervation	Aluminum hydroxide crystals	DPT, MMR Hepatitis B H. influenza (18 mo)	+
7	5 years 10 mo/M	Myoglobinuria. Rhabdomyolysis. Family history of rhabdomyolysis	Phospho-glycerate kinase deficiency (<19%)	Aluminum hydroxide crystals	DPT, MMR Hepatitis B H. influenza (2 y)	+
8	2 y 8 mo/F	Weakness, developmental delay, ischemic infarctions in cerebellum, pons, medulla oblongata. Probable MELAS syndrome	Normal muscle <sup>c</sup>	NA	DPT, MMR Hepatitis B H. influenza (2 y)	+

NOTE: EM = electron microscopy; NA = not available/outside consultation cases; DPT = diphtheria, pertussis, tetanus; MMR = measles mumps rubella; MELAS = myopathy, encephalopathy, lactic acidosis, stroke-like episodes; COX = cytochrome *c* oxidase.

a. All biopsies demonstrated histological features of macrophagic fasciitis.

b. Bilateral thigh injections 2 months to 1 year before biopsy, up to 11 vaccinations, approximate age at the time of last injection.

c. Paraffin sections only.

fibers, without red ragged fibers or apparent abnormalities in NADH dehydrogenase or succinic dehydrogenase. Cytochrome oxidase was normal in cases 2 and 4, and absent in most fibers in case 6. Case 5 showed generalized rounding, size variability, scattered enlarged dystrophic and necrotic fibers, and focal fat ingrowth; it was entirely negative for the carboxyl domain of dystrophin. Case 7 showed minimal variability in the size and shape of muscle fibers and marked type I preponderance (more than 95% of fiber population). Biochemical testing (Women's and Children's Hospital of Buffalo, Buffalo, New York) revealed reduction of phosphoglycerate kinase to 19% of control values. The biopsy in case 8 was available in paraffin-embedded tissue and, except for marked accumulation of macrophages and occasional regenerating fibers, showed no abnormalities.

Electron microscopy showed macrophages with intracytoplasmic, osmiophilic spicular inclusions of aluminum hydroxide in 2 cases (6 and 7; Figure 2). Plastic-embedded

tissue from 4 cases (1, 2, 3, 5) did not contain macrophages, and tissue was not available in 2 consultation cases (cases 4 and 8).

## Discussion

Adults with macrophagic myofasciitis usually report myalgia, arthralgia, and other symptoms reminiscent of chronic fatigue syndrome.<sup>1-4,16,17</sup> The original description of macrophagic myofasciitis<sup>1</sup> as a new entity, as well as the majority of subsequently reported patients originated from France.<sup>2-4,16-22</sup> Because the French patients were previously immunized against hepatitis B (84–86%), hepatitis A (19%), and tetanus (58%) 3–96 months before the biopsy, difficulty in clearing aluminum from the injection site and an abnormal immune response to prolonged tissue retention of aluminum hydroxide were considered to be the etiology of the

disease.<sup>1-4,22</sup> The role of immunity was further stressed by reports of cases associated with a multiple sclerosis-like syndrome,<sup>16</sup> rheumatoid arthritis,<sup>20</sup> systemic lupus erythematosus, and inclusion body myositis.<sup>18</sup> Presence of circulating autoantibodies,<sup>1,3</sup> the frequent association with DRB1 human lymphocyte antigen genotype, and a consistent response to corticosteroids<sup>1,4,13,21</sup> also suggested an immune-mediated syndrome. However, the increased frequency of immunological disorders among patients with myofasciitis can also be explained by selection bias, because many individuals with autoimmune diseases and myalgia undergo muscle biopsy.<sup>23,24</sup> Moreover, despite systemic myalgia, the granulomas are always limited to the injection sites and their presence may represent coincidental focal reaction to aluminum derivatives, unrelated to the underlying systemic or neuromuscular disease.<sup>6,14,23,24</sup> Examples of such fortuitous associations of myofasciitis and other conditions such as centronuclear myopathy,<sup>25</sup> ocular myopathy,<sup>19</sup> and facio-scapulo-humeral dystrophy<sup>5</sup> have been described previously. The selection of the deltoid muscle for vaccination as well as biopsy site in France, may explain the almost exclusive occurrence of this disease in the French population.<sup>23</sup> On the other hand, the virtual absence of this entity in adults from other countries could be related to strict adherence to selection of the biopsy site remote from any previous injections. To the best of our knowledge, no case of myofasciitis has been reported when a muscle other than at the site of vaccination was selected for biopsy in an adult. By analogy, the quadriceps femoris is the site of choice for immunization as well as biopsy in all the reported pediatric cases with this condition. Also, all our patients were vaccinated by multiple, bilateral injections to "thigh muscles," from 2 months to 1 year before the biopsy of the vastus lateralis muscle.

Despite world-wide childhood immunization programs, only few pediatric cases of macrophagic myofasciitis have been published.<sup>6-12,14,15</sup> Rivas et al<sup>7</sup> reported 7 children with this diagnosis out of 490 muscle biopsies from several Spanish centers. Lacson et al<sup>14</sup> described pathological changes consistent with macrophagic myofasciitis in 2 of 220 pediatric muscle biopsies over an 8-year period. These figures are similar to 8 cases in approximately 800 predominantly pediatric biopsies over a 5-year period in our institution. One of the conceivable explanations for the rarity of in routine pediatric biopsies is the small size of macrophage collections—usually below 1 mm in diameter in our samples.

Almost consistently, pediatric patients show poor or no correlation between the clinical presentations and biopsy findings. Among the reported cases of childhood myofasciitis, only 1 had clinical signs of muscle disease and improved after corticosteroid treatment, most likely due to coincidence of myofasciitis and vasculitis.<sup>10</sup> In other reports, myofasciitis was associated with motor delay and hypotonia that was difficult to explain by muscle biopsy findings.<sup>10,12,14</sup> In 1 of these patients, it was considered a cause of motor retardation,<sup>10</sup>

whereas the other reported children with this entity had unrelated medical problems.<sup>6,9,14</sup> Among Arabic children with myofasciitis,<sup>6</sup> 4 exhibited central nervous system disorders and none had muscle disease or responded to corticosteroids. In a recently reported series of 7 pediatric patients from Spain, all had nonrelated neurological disease, most often mitochondrial cytopathies.<sup>7</sup> Five of our patients suffered from a clearly defined neuromuscular disorder (Duchenne muscular dystrophy, cytochrome *c* oxidase deficiency, phospho-glycerate deficiency, and spinal muscular atrophy [in 2 children]). The remaining 3 children with morphologically normal muscle, had a family history and/or clinical manifestations of serious neurological disease with lactic acidosis, consistent with mitochondrial cytopathies.<sup>26</sup> Recently, Nevo et al<sup>6</sup> described macrophagic myofasciitis in 6 Arabic children and suggested that parental consanguinity may play a role in genetic susceptibility for this disease. Five of our patients were products of consanguineous unions. Although unlikely, increased risk of developing an unusual vaccination site reaction due to consanguinity remains an open question, especially in the Arabic population with frequent intermarriages.

Because of small sizes of specimens and sampling error inherent to electron microscopy, characteristic aluminum hydroxide deposits were found only in 2 of our cases. However, the light microscopic and immunohistochemical features were diagnostic of macrophagic myofasciitis. These morphological changes were indistinguishable from postvaccination granulomas in immunized patients<sup>25</sup> and experimental animals.<sup>22,25,27,28</sup> They differed from adverse tissue reaction to intramuscular drug injections<sup>29,30</sup> or undesirable postimmunization site reactions that display acute inflammation with necrosis, microabscesses, eosinophils, and foreign body granulomas.<sup>31-33</sup> Although myonecrosis has not been seen in adults with myofasciitis,<sup>1-4</sup> it was obvious in 2 of our biopsies, most likely due to a greater sensitivity of the intrinsically diseased and immature muscle of children as compared to essentially healthy adults. Muscle necrosis was also observed in 2 of 7 pediatric patients in the Spanish study.<sup>7</sup> However, because regenerating muscle fibers have been reported in this condition,<sup>6,14</sup> including all our cases, the presence or absence of necrosis in the tissue could be a consequence of the time interval between immunization and biopsy. Fiber necrosis in the biopsy of patient with Duchenne muscular dystrophy obviously represented 1 of the morphological manifestations of the disease process.

Significant doubts have been raised about the relevance of characteristic aluminum containing lesions in muscle<sup>6,7,11,14,23-25,28</sup> to clinical manifestations of macrophagic myofasciitis. Magnetic resonance spectroscopy showed no abnormalities in muscle energy metabolism in patients with this disease.<sup>34</sup> Furthermore, experimental injections of vaccines with aluminum hydroxide adjuvants produce persistent lesions identical to that seen in macrophagic

myofasciitis in animals that remain asymptomatic.<sup>28,32</sup> The incidental nature of these granulomas was also documented in a single human autopsy case.<sup>25</sup>

The clinical presentations in our patients cannot be explained by the granulomas in the muscle. Each of the reported children suffered from specific neurological disease proven by either biopsy or supplementary clinical and biochemical studies. We conclude, therefore, that in the so-called childhood macrophagic myofasciitis, the intramuscular aluminum-containing granulomas represent a localized reactive process at the vaccine injection site, rather than an expression of a specific systemic inflammatory disease. Therefore, detection of these lesions in muscle biopsy does not preclude another coexistent muscle disease. We believe that the emergence of macrophagic myofasciitis as an entity is a consequence of inappropriate selection of the previously immunized muscle group for diagnostic biopsy in subjects with concomitant neuromuscular or systemic disease.

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