GYNECOLOGY

Characterization of the host inflammatory response following implantation of prolapse mesh in rhesus macaque

Bryan N. Brown, PhD; Deepa Mani, MBBS; Alexis L. Nolfi, BS; Rui Liang, MD; Steven D. Abramowitch, PhD; Pamela A. Moalli, MD, PhD

OBJECTIVE: We sought to determine the predominant cell type (macrophage, T lymphocyte, B lymphocyte, mast cell) within the area of implantation of the prototypical polypropylene mesh, Gynemesh PS (Ethicon, Somerville, NJ); and to determine the phenotypic profile (M1 proinflammatory, M2 antiinflammatory) of the macrophage response to 3 different polypropylene meshes: Gynemesh PS (Ethicon), and 2 lower-weight, higher-porosity meshes, UltraPro (Ethicon) and Restorelle (Coloplast, Humblebaek, Denmark).

STUDY DESIGN: Sacrocolpopexy was performed following hysterectomy in rhesus macaques. Sham-operated animals served as controls. At 12 weeks postsurgery, the vagina-mesh complex was excised and the host inflammatory response was evaluated. Hematoxylin and eosin was used to perform routine histomorphologic evaluation. Identification of leukocyte ($CD45^+$) subsets was performed by immunolabeling for CD68 (macrophage), CD3 (T lymphocyte), CD20 (B lymphocyte), and CD117 (mast cell). M1 and M2 macrophage subsets were identified using immunolabeling ($CD86^+$ and $CD206^+$, respectively), and further evaluation was performed using enzyme-linked immunosorbent assay for 2 M1 (tumor necrosis factor-alpha and interleukin [IL]-12) and 2 M2 (IL-4 and IL-10) cytokines.

RESULTS: Histomorphologic evaluation showed a dense cellular response surrounding each mesh fiber. $CD45^+$ leukocytes accounted for $21.4 \pm 5.4\%$ of total cells within the perimesh area captured in a $\times 20$ field, with macrophages as the predominant leukocyte subset ($10.5 \pm 3.9\%$ of total cells) followed by T lymphocytes ($7.3 \pm 1.7\%$), B lymphocytes ($3.0 \pm 1.2\%$), and mast cells ($0.2 \pm 0.2\%$). The response was observed to be more diffuse with increasing distance from the fiber surface. Few leukocytes of any type were observed in sham-operated animals. Immunolabeling revealed polarization of the macrophage response toward the M1 phenotype in all mesh groups. However, the ratio of M2:M1 macrophages was increased in the fiber area in UltraPro ($P = .033$) and Restorelle ($P = .016$) compared to Gynemesh PS. In addition, a shift toward increased expression of the antiinflammatory cytokine IL-10 was observed in Restorelle as compared to Gynemesh PS ($P = .011$).

CONCLUSION: The host response to mesh consists predominantly of activated, proinflammatory M1 macrophages at 12 weeks postsurgery. However, this response is attenuated with implantation of lighter-weight, higher-porosity mesh. While additional work is required to establish causal relationships, these results suggest a link among the host inflammatory response, mesh textile properties, and clinical outcomes in the repair of pelvic organ prolapse.

Key words: cytokines, inflammatory response, macrophage phenotype, polypropylene mesh, rhesus macaque

Cite this article as: Brown BN, Mani D, Nolfi AL, et al. Characterization of the host inflammatory response following implantation of prolapse mesh in rhesus macaque. Am J Obstet Gynecol 2015;213:668.e1-10.

More than 250,000 women per year in the United States will undergo surgery for the treatment of pelvic organ prolapse, with direct costs totaling >$1 billion.[1-3] Native tissue repair has a recurrence rate of 40% at 2 years[4,5]; therefore, mechanical reinforcement of tissues using synthetic mesh has increased over the last decade.[6] While

From the Departments of Bioengineering (Drs Brown, Abramowitch, and Moalli and Ms Nolfi) and Obstetrics, Gynecology, and Reproductive Sciences (Drs Brown, Liang, Abramowitch, and Moalli), and McGowan Institute for Regenerative Medicine (Drs Brown, Mani, and Moalli), University of Pittsburgh; and Magee–Womens Research Institute (Drs Liang and Moalli), Pittsburgh, PA.

Received April 15, 2015; revised June 21, 2015; accepted Aug. 2, 2015.

This work was supported by National Institutes of Health awards R01 HD061811 (P.A.M.) and K12HD043441 (B.N.B.). The content is solely the responsibility of the authors and does not necessarily represent the official views of the National Institutes of Health. The funding source had no involvement in the study design, collection of data, analysis of data, interpretation of data, writing of the report, or the decision to submit for publication.

The authors report no conflict of interest.

Presented at the Joint Scientific Meeting of the American Urogynecologic Society and International Urogynecologic Association, Washington, DC, July 22-26, 2014.

Corresponding author: Pamela A. Moalli, MD, PhD. pmoalli@mail.magee.edu

0002-9378/$36.00 • • http://dx.doi.org/10.1016/j.ajog.2015.08.002

TABLE 1
Mechanical and structural characteristics associated with each mesh[12,26]

	Gynemesh PS (Ethicon)	UltraPro (Ethicon)	Restorelle (Coloplast)
Weight, g/m^2	44	31	19
Pore size, μm	2240	$\geq$4000[a]	2370
Porosity, %	64 $\pm$ 2.1	69 $\pm$ 1.8	78 $\pm$ 3.0
Stiffness, N/mm	28 $\pm$ 2.7	22 $\pm$ 2.8	11 $\pm$ 0.89

[a] UltraPro contained resorbable component (poliglecaprolactone 25) in addition to polypropylene allowing it to have very large pores (4 mm) when this component is resorbed; values reported with resorbable component dissolved.

Brown. Inflammatory response to prolapse mesh. Am J Obstet Gynecol 2015.

mesh implantation has been shown to improve anatomical outcomes in the anterior and apical compartments, complications are observed, particularly with transvaginal placement,[7-11] including mesh exposure through the vaginal wall, shrinkage, erosion, and pain.

Recent work suggests that mesh exposures may be induced by stress shielding. That is, a mismatch in stiffness between the mesh and tissue lead to degeneration of the underlying vagina and a loss of mechanical integrity over time. This maladaptive remodeling response precipitates atrophy of the smooth muscle layer associated with a decrease in contractility as well as a shift in tissue extracellular matrix composition and a loss of biomechanical integrity.[12-14] Differences in mesh properties (weight, pore size, porosity, stiffness) were shown to be related to the degree to which this degenerative process occurs, with higher-weight, lower-porosity, and increased-stiffness mesh being associated with increased vaginal tissue degradation. Mesh with higher weight, lower porosity, and increased stiffness has also been suggested to result in increased rates of complications in clinical practice.[15,16]

Mesh complications may also be attributable to the inflammatory processes associated with the macrophage-predominated foreign body reaction mounted by the host following implantation. Without question, the long-term presence of activated proinflammatory cells can have a negative impact on the ability of a material to function as intended. However, a number of recent studies have demonstrated that the macrophage response is also an essential component of the process leading to tissue incorporation, and functional remodeling of implanted materials, suggesting the potential for phenotypic dichotomy in the host response.[17,18] Indeed, macrophages have been classified as having diverse and plastic phenotypes along a continuum between M1 (classically activated; proinflammatory) and M2 (alternatively activated; regulatory, homeostatic) extremes.[19-21] An increasing number of studies in the field of biomaterials have begun to apply these paradigms and concepts, showing that macrophage polarization is a predictor of integration following implantation in multiple applications.[18,22-25] However, the macrophage response following implantation of surgical mesh with varying characteristics has not been described. Moreover, limited studies to date have addressed the impact of mesh implantation on the vagina—an organ with an immunologically distinct environment from that of other tissues in which the host response to mesh has been examined.

The objectives of the present study were 2-fold: (1) to determine the predominant cell type (macrophage, T lymphocyte, B lymphocyte, mast cell) within the area of implantation of the prototypical polypropylene mesh, Gynemesh PS (Ethicon, Sommerville, NJ); and (2) to determine the phenotypic profile (M1 proinflammatory, M2 antiinflammatory) of the macrophage response to 3 different polypropylene meshes: Gynemesh PS, and 2 lower-weight, higher-porosity meshes, UltraPro (Ethicon) and Restorelle (Coloplast, Humblebaek, Denmark).

TABLE 2
Demographic data collected (age, weight, gravidity, and parity)

Groups	Age, y[a]	Parity[b]	Weight, kg[a]	POP-Q stage[b]
Sham	12.6 $\pm$ 2.8	3 (2, 6)	7.3 $\pm$ 1.4[c]	0 (0, 1)
Gynemesh PS	12.9 $\pm$ 2.2	4 (3.8, 5)	8.2 $\pm$ 1.6	0 (0, 0)
UltraPro	13.0 $\pm$ 2.2	3.5 (2, 5.8)	7.8 $\pm$ 1.4	0 (0, 0.25)
Restorelle	13.8 $\pm$ 1.7	5 (3, 5.5)	10.0 $\pm$ 2.8[c]	0.5 (0, 1.3)
P value[d]	.780	.970	.042	.700

[a] Mean $\pm$ SD; [b] Median (first quartile, second quartile); [c] Statistical significance between groups ($P < .05$); [d] Comparison of overall *P* value among groups.

Brown. Inflammatory response to prolapse mesh. Am J Obstet Gynecol 2015.

Materials and Methods

Meshes

The test articles consisted of 3 polypropylene meshes with varying textile and mechanical characteristics as previously described.[12,26] Briefly, specific weight and pore size were provided by the manufacturer. Porosity was determined using a custom-designed algorithm (Matlab, Version 8.0; Mathworks, Natick, MA) and stiffness was determined by ball burst testing. Table 1 shows the relevant mechanical and structural characteristics associated with each mesh. Of note, UltraPro is manufactured with an absorbable component (poliglecaprolactone 25) in addition to polypropylene allowing it to have very large pores (4 mm) when this component is fully absorbed.

Animals

The samples for the present study were obtained from a larger study.[12,13]