Data in Brief

Loss of Drp1 in the liver leads to an alteration in expression of the genes involved in the immune system

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A B S T R A C T

Dynamin-related protein 1 (Drp1) is a member of the dynamin family of large GTPase, which cycles between the cytosol and the mitochondrial outer membrane, and mediates mitochondrial fission. Using microarray analysis of gene expression in the livers of wild-type and Drp1 knockout mice, we have previously identified that endoplasmic reticulum (ER) stress marker genes are significantly increased by the absence of Drp1 [1]. Here, we provide methodological and analytical details of the microarray data, which have been deposited in the Gene Expression Omnibus as data set GSE64222. We have performed further gene ontology analysis of the data and found the differential expression of a subset of genes that are involved in the immune response in the livers of Drp1 knockout mice versus wild-type controls.

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1. Direct link to deposited data

The deposited data can be found at: http://www.ncbi.nlm.nih.gov/geo/query/acc.cgi?acc=gse64222.

2. Experimental design, materials and methods

Mitochondria are highly dynamic organelles that frequently fuse and divide in disease, aging, and development [2]. In vertebrates, mitofusin-1 and -2 (MFN1 and MFN2) are involved in mitochondrial fusion, whereas DRP1 and mitochondrial fission factor (MFF) control mitochondrial fission [3]. Recently, it was reported that the ER plays an active role in defining the sites of mitochondrial division [4]. In fact, mitochondria and the ER physically interact by close structural juxtaposition, via the mitochondrial-associated ER membrane (MAM). To clarify the role of mitochondrial fission in this communication, we generated mice lacking the mitochondrial fission protein Drp1 in the liver (Drp1LKO). When the mice were fed with a high-fat diet (HFD), analysis of gene expression in the liver demonstrated marked elevation of ER stress markers. We also found a second subset of genes, discussed here, that are involved in the immune response.

2.1. Animals

Drp1LKO mice were generated by crossing Drp1floxed/+ mice with Alb-Cre mice [5]. Mice were fed ad libitum with a normal chow diet (5.4% fat, CRF-1, Orient Yeast Co., Tokyo, Japan) and kept under a light–dark cycle of 12 h. For the HFD study, 4-week-old mice were put on high-fat diet (24% fat, lard fat, 45 kcal % fat, D12451; Research Diets, New Brunswick, NJ) for 24 weeks. All mouse procedures and protocols were in accordance with the Guide for the Care and Use of Laboratory Animals and were approved by the Ethics Committees on Animal Experimentation (Kyushu University, Graduate School of Medicine).

2.2. RNA isolation and microarray

After 24 weeks high fat diet, mice were fasted for 17 h and then sacrificed. Total RNA was isolated from cells using TRIzol Reagent (nitrogen) and purified using SV Total RNA Isolation System (Promega).
cRNA was amplified and labeled using a Low input Quick Amp Labeling Kit (Agilent Technologies). cRNA was hybridized to a 44K 60-mer oligomicroarray (Whole Mouse Genome Microarray 4 × 44K v2; Agilent Technologies) according to the manufacturer’s instructions. The hybridized microarray slides were scanned using an Agilent scanner. The relative hybridization intensities and background hybridization values were calculated using Feature Extraction Software version 9.5.1.1 (Agilent Technologies). The scanned images were analyzed with Feature Extraction Software 9.5.1.1 (Agilent) using default parameters to obtain background subtracted and spatially detrended Processed Signal intensities. The raw signal intensities and flags for each probe were calculated from the hybridization intensities and spot information according to the procedures recommended by Agilent Technologies using the Flag criteria in the GeneSpring Software.

3. Results

Genes were selected using the criterion of a Z score of ≥ 2, which identified 526 up-regulated and 640 down-regulated genes in Drp1LiKO mice. The top 5 up-regulated genes have been reported in other publications, of these, 3 genes are known to be ER stress response genes, which was the topic of our previously published article [1]. To further investigate these data, we use the DAVID tool to functionally cluster up-regulated and down-regulated genes by similarly annotated gene ontology (GO) biological process terms, respectively. The top ten significantly enriched annotation clusters of up-regulated genes were shown in Fig. 1. We found that seven of the ten clusters were related to the immune system; these clusters included terms such as immune response, phagocytosis, antigen processing and presentation, defense response and response to virus. The remaining clusters were related to 2'-deoxyribonucleotide biosynthetic process, amine biosynthetic process and cell death. Next, to further define connections between immune molecules that were regulated by mitochondrial fission, we list the 32 genes selected by using the term “immune response” in Table 1. The most significant ten clusters for down-regulated genes were presented in Fig. 2, and include lipid biosynthetic process, fatty acid metabolic process, acute inflammatory response, actin filament-based process, cell fate commitment, response to wounding, actomyosin structure organization, complement activation, alternative pathway, actin cytoskeleton organization and brown fat cell differentiation.

4. Discussion

We described here that lack of DRP1 increased the expression levels of a large number of genes involved in immune response. Within the past several years, a couple of studies have shed light on the connection between mitochondrial dynamics and antiviral innate immunity [6]. Indeed, Castanier and colleague have observed that knockdown of Drp1 increased innate immune signaling and they have highlighted the importance of the interconnectedness and interdependence of mitochondria fission in antiviral innate immunity in vitro [7]. In this study, we provided the compelling evidence that mitochondrial fission regulates the expression of the genes responsible for the immune system in the liver.

Conflict of interest

The authors declare no conflict of interests.

Acknowledgments

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**Fig. 1.** Microarray functional annotation summary results of the up-regulated genes.
Table 1
Genes involved in the immune response.

| Gene symbol | Description                                      | WT signal | KO signal | Z score | Ratio | GenBank accession |
|-------------|--------------------------------------------------|-----------|-----------|---------|-------|------------------|
| Oas1a       | 2′-5′-oligoadenylate synthetase 1A               | 1602      | 10,770    | 5.41    | 6.72  | NM_145211        |
| Oas1f       | 2′-5′-oligoadenylate synthetase 1F               | 719       | 4818      | 5.40    | 6.70  | NM_145153        |
| Apo4        | Apolipoprotein A-IV                              | 8937      | 34,821    | 4.83    | 3.90  | NM_007468        |
| Cxcl10      | Chemokine (C-X-C motif) ligand 10                | 817       | 3872      | 4.36    | 4.74  | NM_021274        |
| Rad52       | Radical S-adenosyl methionine domain containing 2| 915       | 4104      | 4.27    | 4.49  | NM_021384        |
| Ccl5        | Chemokine (C-C motif) ligand 5                   | 834       | 3786      | 4.24    | 4.54  | NM_013653        |
| Defb1       | Defensin beta 1                                  | 56        | 341       | 4.17    | 6.14  | NM_007843        |
| Oas2l       | 2′-5′-oligoadenylate synthetase-like 2           | 277       | 1088      | 3.83    | 3.92  | NM_011854        |
| Oas2        | 2′-5′-oligoadenylate synthetase 2                | 400       | 1485      | 3.68    | 3.72  | NM_145227        |
| Irf7        | Interferon regulatory factor 7                   | 829       | 2906      | 3.52    | 3.50  | NM_016830        |
| Igh-1a      | Immunoglobulin heavy chain 1a                    | 312       | 1042      | 3.38    | 3.34  | AK007918         |
| Clec7a      | C-type lectin domain family 7, member a          | 645       | 2131      | 3.35    | 3.31  | NM_020008        |
| Sqstm1      | Sequestosome 1                                   | 3817      | 12,111    | 3.28    | 3.17  | NM_011018        |
| Oas3        | 2′-5′-oligoadenylate synthetase 3                | 24        | 332       | 3.08    | 13.65 | NM_145226        |
| Oas1l       | 2′-5′-oligoadenylate synthetase-like 1           | 3388      | 9930      | 3.06    | 2.93  | NM_145209        |
| H2-D1       | Histocompatibility 2, D region locus 1           | 34,888    | 65,382    | 2.72    | 1.87  | NM_010380        |
| H2-Q2       | Histocompatibility 2, Q region locus 2           | 32,901    | 59,740    | 2.59    | 1.82  | NM_010392        |
| Ccl14       | CD14 antigen                                     | 175       | 541       | 2.58    | 3.08  | NM_009841        |
| H2-K1       | Histocompatibility 2, K1, K region, transcript variant 1 | 30,325 | 54,943 | 2.58 | 1.81 | NM_001001892 |
| Ccl7        | Chemokine (C-C motif) ligand 7                   | 12        | 101       | 2.57    | 8.78  | NM_013654        |
| Gbp3        | Guanylate binding protein 3                      | 593       | 1450      | 2.51    | 2.45  | NM_018734        |
| Raet1e      | Retinoic acid early transcript 1E                | 1264      | 3025      | 2.48    | 2.39  | NM_198193        |
| C1rb        | Complement component 1, r subcomponent B         | 813       | 1853      | 2.31    | 2.28  | NM_001113356     |
| H2-T23      | Histocompatibility 2, T region locus 23         | 39,824    | 67,056    | 2.26    | 1.68  | NM_010398        |
| Icosl       | ICOS ligand precursor (B7 homolog 2) (B7-H2)     | 6         | 126       | 2.25    | 21.20 | AF394451         |
| Ccl2        | Chemokine (C-C motif) ligand 2                   | 204       | 543       | 2.24    | 2.66  | NM_011333        |
| Mpa2l       | Macrophage activation 2 like                     | 512       | 1127      | 2.21    | 2.20  | NM_194336        |
| Ccl4        | Chemokine (C-C motif) ligand 4                   | 88        | 221       | 2.11    | 2.51  | NM_013652        |
| Spon2       | Spondin 2, extracellular matrix protein          | 987       | 2080      | 2.09    | 2.11  | NM_133903        |
| Gm111127     | T16 class I MHC gene (exon 5) fragment           | 21,436    | 38,601    | 2.09    | 1.80  | AB359227         |
| LOC676689    | Similar to H-2 class I histocompatibility antigen, L-D alpha chain precursor | 8739 | 15,605 | 2.06 | 1.79 | XM_992161 |
| Igh-VJ358    | Immunoglobulin heavy chain V gene segment        | 6         | 97        | 2.01    | 16.02 | AF296435         |

Fig. 2. Microarray functional annotation summary results of the down-regulated genes.
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