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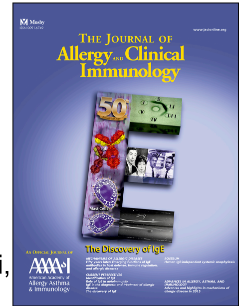

Pulmonary Sarcoidosis is Associated With Exosomal Vitamin D-Binding Protein and Inflammatory Molecules

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PULMONARY SARCOIDOSIS IS ASSOCIATED WITH EXOSOMAL VITAMIN D-BINDING PROTEIN AND INFLAMMATORY MOLECULES

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AUTHOR CONTRIBUTION

Conceived and designed the experiments: S.G., K.R.Q., J.G., A.E., C.J.E.W and R.M. Performed the experiments: K.R.Q., R.M., M.J.M.B., C.J.E.W and A.L. Analyzed the data: M.J.M.B., R.M. and C.J.E.W. Recruited patients and samples: S.G., J.G. and A.E. Wrote the paper, all authors.

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30 **ABSTRACT**

31 *BACKGROUND*: Sarcoidosis is an inflammatory granulomatous disorder characterized by
32 accumulation of Th-1 type $CD4^+$ T cells and immune-effector cells within the affected organs,
33 most frequently the lungs. Exosomes are extracellular vesicles conveying intercellular
34 communication, with possible diagnostic and therapeutic applications.
35 *OBJECTIVES*: We have aimed to provide an understanding of the pro-inflammatory role of
36 bronchoalveolar lavage fluid (BALF) exosomes in sarcoidosis, and to find candidates for
37 disease biomarkers.
38 *METHODS*: We performed a mass spectrometric proteomics characterization of BALF
39 exosomes from 15 sarcoidosis patients and 5 healthy controls, and verified the most
40 interesting results with flow cytometry, ELISA and western blot analyses in an additional 39
41 patients and 22 controls.
42 *RESULTS*: More than 690 proteins were identified in the BALF exosomes, several of which
43 displayed significant upregulation in patients, including inflammation-associated proteins
44 such as Leukotriene A_4 Hydrolase. Most of the complement-activating factors were
45 upregulated, whereas the complement-regulatory CD55 was lower in patients compared to
46 healthy controls. In addition, we detected for the first time Vitamin D binding protein (VDBP)
47 in BALF exosomes, which was more abundant in patients. To evaluate exosome-associated
48 VDBP as a biomarker for sarcoidosis, we investigated plasma exosomes from 23 patients and
49 11 healthy controls and found significantly higher expression in patients.
50 *CONCLUSION*: Together, these data contribute to understanding the role of exosomes in lung
51 disease, and provides suggestions for highly warranted sarcoidosis biomarkers. Further, the
52 validation of an exosome-associated biomarker in the blood of patients opens up for novel,
53 and less invasive, support for disease diagnosis.
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55 **KEYWORDS**

56 Exosomes, extracellular vesicles, sarcoidosis, leukotrienes, Vitamin D-binding protein,
57 proteome, biomarkers, complement.

58 **CLINICAL IMPLICATIONS**

59 Sarcoidosis is associated with invasive diagnostic procedures, and exosomes represent
60 potential new diagnostic and prognostic biomarkers. These data also contribute to the
61 understanding of the inflammatory properties of exosomes.

62 **CAPSULE SUMMARY**

63 Our clinical data resulting from proteomic analysis of BALF exosomes, followed by multiple-
64 technique validations contribute to understanding how exosomes contribute to inflammation
65 in airway disease, and suggest novel diagnostic and prognostic markers for Sarcoidosis.

66 **INTRODUCTION**

67 Sarcoidosis is an inflammatory granulomatous disorder, affecting mostly the lungs.
68 Spontaneous remission often occurs, although around one-third of the cases develop chronic
69 disease, which may be fatal [1, 2]. The etiology of sarcoidosis is not yet fully understood, but
70 immunologic evidence and its geographical variation have suggested causes including
71 infection, occupational exposure, as well as genetic factors. The presence of clonal Th1-like
72 CD4+ T cells, macrophages, and immune-effector cells within affected organs suggests an
73 antigen-driven autoimmune disease [3]. Autoantigens, such as vimentin and ATP-synthase,
74 have been identified as targets for expanded T cell clones in the lung [4], and other data
75 suggest roles of microorganisms, on account of the T cell reactivity against mycobacteria-
76 derived mKatG [5].

77 Exosomes are 30-150 nm vesicles derived from endosomal compartments, which act as
78 messengers between cells [6] and can either stimulate or inhibit immune cells, depending on
79 their cellular origin [7]. Dendritic cell exosomes can stimulate T cells [8, 9], and are
80 promising cancer vaccine candidates [10, 11], but exosomes from the gut and cancer cells
81 seem to inhibit the immune system [12, 13]. Exosomes have been found in most body fluids
82 including breast milk [14] and plasma [15], and are likely to play physiological roles, and
83 have potential as disease biomarkers. We have previously found exosomes in bronchoalveolar
84 lavage fluid (BALF) of healthy individuals [16], and shown in functional studies that
85 sarcoidosis patient BALF is enriched in pro-inflammatory exosomes and can induce
86 production of interferon gamma and interleukin-8 *in vitro* [17]. In further functional studies,
87 we have shown that macrophage and dendritic cell-derived exosomes carry leukotriene
88 pathway components and have migration-inducing capacities [18], suggesting that exosomes
89 may contribute to airway remodelling. We have also established that BALF exosomes in
90 asthmatic patients carry leukotriene-forming enzymes and functionally induce production of
91 cytokines and leukotrienes [19]. Recently, it was also found that exosomes promote the
92 chemotaxis of neutrophils by transporting leukotriene B4 [20]. Taken together, the
93 accumulating evidence points to a role of exosomes in leukotriene-mediated intercellular
94 communication, possibly with implications in airway disease.

95 In view of the latter evidence, we aimed at thoroughly dissecting disease-associated
96 components of BALF exosomes from sarcoidosis patients. We conducted a full proteomic
97 analysis of the exosomal contents, followed by validations of the most interesting results
98 using flow cytometry, ELISA, Western blot and nanoparticle tracking analysis. We detected
99 clear proteomic differences between exosomes from sarcoidosis patients and controls, with
100 pro-inflammatory and immune system activation pathway components more abundant in
101 patient exosomes. Several of the proteomic findings, including an increase in leukotriene-

102 forming enzymes, were successfully validated by several techniques, as were a number of
103 potential biomarkers. These findings indicate the importance of future investigations of the
104 role of these proteins on exosomes in sarcoidosis, but also as disease biomarkers to support
105 the invasive and circumstantial diagnostic procedures of sarcoidosis patients.

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107 **MATERIALS AND METHODS**

108 **Study subjects**

109 BALF from 44 sarcoidosis patients (median age 46, 52% males, and 11% smokers), three
110 fibrosis patients, two asthma patients and one alveolitis patient was obtained as part of routine
111 diagnostic investigations at Karolinska Hospital, Sweden. Sarcoidosis diagnoses were
112 established according to WASOG guidelines [21], based on typical clinical signs, biopsies
113 showing non-caseating granuloma formation, and chest radiographic findings compatible with
114 sarcoidosis. The diagnoses were further supported by differential BALF cell counts, BALF
115 CD4/CD8 ratio and by ruling out other causes of these observations. Patients with Löfgren´s
116 syndrome (n=9) were identified by acute onset of the disease with fever, erythema nodosum,
117 and/or ankle arthritis, and bilateral hilar lymphomas with or without concomitant
118 parenchymal infiltrates. None of the patients had received any kind of immunosuppressant
119 therapy at the time of bronchoscopy. Healthy controls (n=22, median age 27, 68% males, and
120 14% smokers) free of medication, with normal chest X-ray, blood cell counts and electrolytes
121 volunteered for BAL. No participants showed signs of respiratory infection at least four
122 weeks prior to the bronchoscopy. All the subjects showed normocalcemia at the time of
123 bronchoscopy, which is reported together with the other characteristics of the patients,
124 including smoking habits, X-ray stages, serum calcium and ACE levels, where available (See
125 Table E1 in this article´s Online Repository at www.jacionline.com). All subjects gave
126 informed consent adhering to protocols approved by the regional ethics committee.

127 **Exosome isolation from BALF and blood**

128 BAL was performed as described previously [22]. The BALF was strained through a double
129 layer of Dacron nets (Millipore, Bedford, Ireland), and centrifuged at 400g for 10 min at 4°C.
130 Cell viability was determined by Trypan blue exclusion and was always >90%. BALF

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131 exosomes were isolated by differential centrifugation as described elsewhere [17]. Plasma
132 exosomes were isolated by centrifuging whole blood at 600g for 10 min, the supernatant was
133 further centrifuged at 20,000g for 20 minutes. This supernatant was filtered through a 0.22µm
134 filter and ultracentrifuged at 140,000g for 90 minutes, the pellet was then washed in PBS.

135 **Sample preparation and iTRAQ labeling**

136 Isobaric tags for relative and absolute quantification (iTRAQ) reagents and buffers were
137 obtained from AB Sciex. Additional reagents, including triethyl ammonium bicarbonate
138 (TEAB), LC-MS grade acetonitrile and methyl methanethiosulfonate (MMTS) were obtained
139 from Sigma. Sequence grade trypsin was used for protein digestion (Promega, Madison, WI,
140 USA). All aqueous solutions were prepared using water from a MilliQ-water purification
141 system. BALF exosomes were prepared for proteomic analysis as previously described [14],
142 with modifications to accommodate for labeling with 8-plex iTRAQ reagents (ABSciex), as
143 described in Online Repository including sample labelling (see Table E3 in this article´s
144 Online Repository at www.jacionline.com).

145 **LC-MS/MS**

146 An Orbitrap-Velos coupled with an EasyNano-LC (Thermo Scientific) was used for LC-
147 MS/MS. Additional analyses were made with QSTAR-Elite (AB Sciex) together with an
148 Ultimate 3000 capillary LC (Dionex). The data were directly analyzed using ProteinPilot™
149 software and the Paragon™ identification algorithm with a human Swiss-Prot database
150 (Release 18/08/2011, 20245 entries).

151 **Flow cytometry**

152 To characterize the exosomes based on original BALF volume, 4.5 µm anti-human HLA-DR
153 beads (Clone HKB-1, Dynal®, Oslo, Norway) were coated with BALF exosomes

154 corresponding to 6 ml of the original BALF volume per µl beads, and stained with FITC-
155 conjugated antibodies to HLA-DR, HLA-ABC, CD9, CD54, CD63, CD81, CD86, DC-SIGN
156 (BD Biosciences, San Diego, CA, USA) or MUC-1 (Abcam Ltd, Cambridge, MA, USA), as
157 previously described [17].

158 For validation of the iTRAQ results, where exosomal quantities were based on total protein
159 contents, 2 µg of BALF exosomes were added per µl of 4 µm latex beads (Molecular Probes,
160 Paisley, UK) coated with anti-human CD63 (BD Biosciences, San Diego, CA, USA) and
161 stained with HLA-DR-FITC, CD63-PE, CD55-FITC (BD Biosciences) or C3-PE (LSBio
162 LifeSpan Bioscience, Inc., Seattle, WA, USA) or with isotype-matched controls. Samples
163 were analyzed by a BD Biosciences FACSCalibur™ and FlowJo software (TreeStar, San
164 Carlos, CA).

165 **Western blot for leukotriene pathway proteins and complement C3**

166 Methods for Western blots (WB) for detection of leukotrienes and complement C3 are found
167 in the Online Repository (see detailed methods in this article´s Online Repository at
168 www.jacionline.com).

169 **ELISA for Vitamin D-binding protein**

170 To quantify exosome-associated VDBP, BALF exosomes from 14 patients, 13 healthy
171 controls and 6 non-sarcoidosis lung disorder patients (3 with fibrosis, 2 asthma, 1 alveolitis)
172 were analyzed using an ELISA kit according to the manufacturer's protocols (R&D Systems,
173 Minneapolis, MN, USA). Both plasma and exosomes isolated from the plasma of 23 patients
174 and 11 healthy controls were analyzed in the same manner.

175 **Sucrose gradient separation of exosomes**

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176 Since VDBP has not been reported associated with exosomes before, we confirmed this by
177 analysing VDBP in exosome fractions isolated by sucrose gradient centrifugation. BALF
178 exosomes from 5 sarcoidosis patients were pooled and separated on a continuous sucrose
179 gradient (0.25-2 mM sucrose and 20 mM HEPES/NaOH, pH 7.4) (Sigma-Aldrich, St Louis,
180 MO, USA), by centrifugation for 20 h at 17,900*g* at 4ºC. Fraction densities were determined
181 by refraction index measurements. Each fraction was ultracentrifuged at 100,000*g* for 35 min
182 to pellet the exosomes for detection of VDBP by ELISA, as described above.

183 **Nanoparticle tracking analysis**

184 To relate the quantities of exosomes between patients and healthy individuals, nanoparticle
185 tracking analysis (NTA) was used, for method description see detailed methods in this
186 article´s Online Repository at www.jacionline.com.

187 **Bioinformatics and statistical analyses**

188 The Reproducibility-Optimised Test Statistic (ROTS) [23] was initially used to analyse the
189 iTRAQ determined protein abundances between the healthy subjects and Sarcoidosis patients.
190 To identify differences between groups, either non-parametric Mann-Whitney test or Kruskal-
191 Wallis were performed using GraphPad Prism 5 (GraphPad Software, Inc. La Jolla, CA,
192 USA). $P<0.05$ was considered significant. More details are found in the Online Repository
193 (detailed methods are found in this article´s Online Repository at www.jacionline.com).

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196 **RESULTS**

197 **Isolation and characterization of exosomes from BALF**

198 In agreement with our previous findings [17], sarcoidosis BALF displayed total exosomal
199 protein amounts greater than two times higher than BALF from healthy controls (p<0.0001)
200 (data not shown). Smoking status did not significantly affect the exosome protein amounts.
201 Patient BALF cell phenotypes are displayed in Figure E3 in this article´s Online Repository at
202 www.jacionline.com.

203 **Phenotype of exosomes analyzed in iTRAQ**

204 Exosomes from the 15 patients and 5 controls included in the iTRAQ analysis were coated to
205 anti-MHC class II beads and analyzed by flow cytometry. These analyses detected the CD63
206 and CD9 exosome markers (Figure 1A-B), as well as HLA-ABC (see Figure E1b in this
207 article´s Online Repository at www.jacionline.com), on both patient and healthy control
208 exosomes. The patient exosomes displayed significantly higher expressions of HLA-DR
209 (p=0.0012) and CD54 (p=0.0052) (Figure 1C-D) compared to the healthy controls. This
210 difference remained significant also when the Löfgren's syndrome patients were excluded
211 from the statistical analysis

212

213 **Identification of proteins in BALF exosomes by iTRAQ**

214 The proteomic profiles of exosomes from 15 patients and 5 healthy controls were analyzed by
215 LC-MS/MS after labelling with iTRAQ reagents. On average 1400 proteins and 16,000
216 peptides were detected in each of the three iTRAQ experiments performed, with the overall
217 detection of 1479 proteins with two or more distinct peptides, at an estimated false discovery
218 rate of 5 % (see this article´s Online Repository at www.jacionline.com). Although 840

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219 proteins were detected in the exosome samples from all subjects (>1 peptide)
220 (http://www.microvesicles.org [24]), we focused on a shorter list of 690 proteins for the
221 statistical analysis, which was filtered to limit the influence of quantification measurements
222 based on single values (See this article´s Online Repository at www.jacionline.com).

223 To gain insight into the biological processes associated with the major changes in protein
224 abundance, the DAVID bioinformatics tool was used. Enrichment and clustering analysis was
225 made for the GO biological processes associated with the proteins altered more than two-fold
226 between the patients and healthy controls. The complete list of GO biological processes are
227 listed (see this article´s Online Repository at www.jacionline.com) and summarized in Figure
228 2. Also, the list of proteins altered two-fold or more between patients and healthy controls are
229 displayed (see Tables E2a and E2b in this article´s Online Repository at
230 www.jacionline.com).

231
232 A range of proteins were more abundant in patient exosomes, including glycoproteins
233 involved in cell-cell interaction, lipopolysaccharide- and Vitamin D-binding protein (VDBP).
234 Kininogen-1, lactotransferrin, prothrombin, serotransferrin, haptoglobulin and apolipoprotein
235 B-100 were also increased in the patients. Patient exosomes further had a higher abundance of
236 proteins involved in homeostasis and hemostasis, as well as inflammation, including
237 membrane attack complex (MAC) proteins and enzymes involved in leukotriene metabolism.
238 The four IgG subclasses and proteins related to activation of adaptive immunity were
239 upregulated in patients. Hierarchical clustering of the iTRAQ results also highlighted the
240 increase in complement system activators (Figure 3), including C3 in patients compared to
241 controls ($p=0.0012$). Reversely, the abundance of complement decay-accelerating factor
242 (CD55), a complement system inhibitor, was higher in the healthy controls ($p=0.0045$)

243 (Figure 4A). Amongst the proteins less abundant in patient exosomes were components from
244 metabolic processes, such as ubiquitination and proteasome activation.

245 **Validation of iTRAQ results**

246 For the proteins highlighted in the iTRAQ analysis, a number of the most promising
247 biomarker candidates and functionally interesting proteins were verified using other
248 techniques. These targets included complement-related proteins, components of the
249 leukotriene pathway and Vitamin D-binding protein.

250 *Complement-related proteins*

251 Flow cytometric analysis of C3 and CD55 on BALF exosomes from 12 patients and 9 healthy
252 controls showed that the lower levels of CD55 seen by iTRAQ (Figure 4A) could be verified
253 (Figure 4B, $p=0.0016$). Complement component C3, however, which was found by iTRAQ to
254 be more abundant in patients (Figure 5A), showed no difference ($p=0.456$) (Figure 5B).
255 However, as flow cytometric analysis detects only epitopes exposed on exosomal surfaces,
256 western blot was used to investigate total C3 levels. The WB analysis showed that C3 was
257 clearly present in patient exosomes, whilst barely detectable in healthy controls (Figure 5C).

258 *Proteins of the leukotriene pathway*

259 LTA_4 hydrolase (LTA_4H) is a potent chemo-attractant for several types of leukocytes and the
260 enzyme that converts LTA_4 to LTB_4. The iTRAQ data indicated that LTA_4H was more
261 abundant in patient exosomes (Figure 6A), and the WB analysis clearly verified this (Figure
262 6B), showing a significantly higher band intensity in the sarcoidosis BALF exosomes
263 compared to healthy exosomes ($p=0.02$, Figure 6C). On the other hand, 5-lipoxygenase (5-
264 LO), which catalyses formation of LTA_4 from arachidonic acid, was found to be 1.3-fold

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265 more abundant in healthy BALF exosomes by iTRAQ (see this article´s Online Repository at
266 www.jacionline.com), which was validated with WB analysis (p=0.024) (Figure 6B).

267 In keeping with our earlier findings [18] we demonstrated a more stable fluorescent signal in
268 5-lipoxygenase-activating protein (FLAP) exosomes compared to β-actin, suggesting FLAP
269 to be a more suitable reference protein for BALF exosomes.

270 *Vitamin D-binding protein*

271 Vitamin D dysregulation has been described in sarcoidosis, and we here present the first
272 evidence associating exosomes to this topic. The iTRAQ analysis indicated a 2-fold
273 upregulation of VDBP (see Table E2a in this article´s Online Repository at
274 www.jacionline.com) in patient BALF exosomes compared to healthy individuals (p=0.0012)
275 (Figure 7A). To validate these differences, we analyzed BALF exosomes from 14 sarcoidosis
276 patients, 13 controls and 6 patients with other lung diseases (three with fibrosis, two with
277 asthma, one with alveolitis) by ELISA. We found significantly higher levels of VDBP in
278 sarcoidosis patient exosomes compared to both healthy controls (Figure 7B) and the patients
279 with lung disorders other than sarcoidosis (data not shown). To further evaluate VDBP as a
280 disease biomarker, we used ELISA to analyze VDBP in plasma from 23 patients and 11
281 controls, but found slightly lower levels of VDBP in patients (Figure 7C). However, when
282 investigating exosomes isolated from the plasma, a significantly higher level of VDBP was
283 seen in the patients (Figure 7D), suggesting that disease-associated levels of VDBP are more
284 distinct in exosomes compared to in whole plasma. This difference remained significant when
285 the Löfgren's syndrome patients were excluded from the statistical analysis.
286 Receiver operating characteristics (ROC) analysis of the plasma
287 exosome-associated levels of VDBP gave an area under curve (AUC) of 0.83 (95%
288 confidence interval of 0.69 to 0.97, p = 0.0024). On the basis if these data, if the

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289 exosomal VDBP concentration would be used as a diagnostic test, a threshold
290 concentration of 24.4 ng/ml, (based on the Youden index of the ROC curve),
291 could be used to discriminate patients from healthy subjects with a sensitivity
292 of 73.9% and a specificity of 81.2%, with positive and negative predictive
293 values of 89% and 60%, respectively (see figure E5 in this article´s Online Repository at
294 www.jacionline.com). However, further measurements from a wider cohort of cases and
295 controls would be needed to robustly validate VDBP as a clinically applicable biomarker for
296 sarcoidosis. We also verified that VDBP is not simply co-pelleted with exosomes during
297 centrifugation by detecting VDBP in the sucrose gradient-fraction corresponding to the
298 exosomal density (see figure E2a in this article´s Online Repository at www.jacionline.com).
299 Plasma exosome quantities were also compared between patients and healthy controls by
300 nanoparticle tracking analysis (NTA), which showed no significant difference in vesicle
301 concentrations between the groups (see figure E2b in this article´s Online Repository at
302 www.jacionline.com). Plasma exosome levels of VDBP were also plotted against disease
303 stage and the main BAL cell composition as well as spirometric data, however showing no
304 correlations (see figure E4 in this article´s Online Repository at www.jacionline.com).

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307

308

309 **DISCUSSION**

310 We have previously demonstrated in functional studies that BALF exosomes from sarcoidosis
311 patients induce pro-inflammatory effects in PBMC and epithelial cells [17], and that asthma
312 patients BALF exosomes when added to epithelial cells induce the production of IL-8 and
313 leukotrienes [19]. To investigate possible mechanisms behind this pro-inflammatory role of
314 exosomes in airway disease, we conducted the current study. We compared the proteome of
315 BALF exosomes between sarcoidosis patients and healthy individuals by iTRAQ to screen for
316 targets for further investigation. The most interesting results were verified by flow cytometry,
317 ELISA and WB. These novel data demonstrate, for the first time, that VDBP is present on
318 BALF exosomes, and significantly increased in sarcoidosis compared to healthy controls. A
319 higher level of VDBP was also found in exosomes isolated from the plasma of sarcoidosis
320 patients (23 patients vs. 11 healthy controls), strengthening the possibility of using exosome-
321 bound VDBP as a biomarker. We also show that exosomes from sarcoidosis patients carry
322 leukotriene-forming enzymes, which is in keeping with our and others' observations of
323 exosome involvement in leukotriene pathways [17-20]. Complement factors and proteins
324 involved in inflammatory processes were also increased in patients, as were proteins involved
325 in innate and adaptive immune responses, protein maturation, hemostasis and homeostasis,
326 which supports a pro-inflammatory role for exosomes in sarcoidosis and may explain their
327 cytokine-inducing capacity, which we previously reported [17].

328

329 Previous studies have shown proteome profiles of total BALF, of lung disorder patients [25-
330 28], including sarcoidosis [29], but not BALF exosomes. In 2002, Magi *et al*. demonstrated
331 that the protein composition of BALF in sarcoidosis patients has a more inflammatory protein
332 profile, including calgranulin A and B, cyclophilin A, complement activators components and
333 VDBP, when compared to idiopathic pulmonary fibrosis (IPF) patients [25]. In addition,

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334 Sabounchi-Schütt *et al*. reported that sarcoidosis BALF samples had higher levels of proteins
335 involved in immune responses and in inflammatory processes compared to healthy controls
336 [26]. However, in our study, which to our knowledge is the first full proteomic analysis with
337 validations of exosome proteins in BALF, the data suggest that exosomes may be more
338 specific markers compared to the whole proteome. Complement activators, such as
339 complement factor B, C1q, C1s, C1r, C2, C3, and membrane attack complex (MAC), showed
340 higher relative abundance in patients, whereas complement inhibitory CD55 and CD59 were
341 decreased. This may reflect a protective role of exosomes, which clear complement activators
342 from cells in inflamed areas by removal of the MAC, possibly to protect surrounding cells
343 from MAC attack [30, 31]. On the other hand, C3-fragment-containing exosomes may
344 potentiate antigen-specific immune responses [32]. Taken together, the presence of
345 complement activators and the low expression of inhibitors of the complement system suggest
346 that BALF exosomes in sarcoidosis patients may play a modulatory role in the disease. The
347 iTRAQ data indicated that the levels of C3 were more than two-fold higher in patients
348 compared to healthy controls. Flow cytometry of the exosomes did not show the same trend,
349 possibly due to a localization of C3 in exosomes inaccessible to flow cytometry antibodies.
350 Indeed, WB analyses for C3 showed that whilst C3 was clearly present in the BALF
351 exosomes from patients, it was barely detectable in the healthy controls, confirming the
352 iTRAQ findings. This could be interpreted as that C3 is only accessible to recipient cells after
353 internalization of exosomes. Intracellular C3 has been shown to sustain T cell homeostasis
354 and mediate effector differentiation [33], and might therefore drive T cell activation and IFN-
355 γ production in these patients. The localization inside exosomes also suggests that C3 is
356 loaded in exosomes during formation and not bound to them after production and release.
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359 LTs are inflammatory mediators produced by oxidation of arachidonic acid through an
360 enzymatic cascade initiated by 5-LO, mediating the epoxide formation by the LTA_4 synthase
361 activity [34]. The final product, LTA_4, can be converted by LTA_4H to proinflammatory
362 leukotriene B_4 [35]. Earlier studies have reported the presence of soluble LTA_4H in human
363 BALF with LTB_4-producing capacities [36], and we have detected LTA_4H in BALF
364 exosomes from allergic asthmatic patients, while 5-LO levels were almost undetectable [19].
365 In the present study, both the iTRAQ results and the western blot analyses showed that
366 LTA_4H was significantly more abundant in sarcoidosis BALF exosomes, while 5-LO was
367 more abundant in controls. This apparently inverted pattern in patients compared with healthy
368 individuals suggest an altered LT pattern in sarcoidosis, and warrants further investigations,
369 especially given the amplitude of already available LT modifying drugs, possibly beneficial
370 for sarcoidosis patients. Our previous findings of IL-8 induction by LT enzymes in asthma
371 exosomes, and the observation of high levels of exosomes in sarcoidosis [17 and this study]
372 suggests that these enzymes associated to exosomes may be conveying inflammatory signals
373 also in sarcoidosis.

374

375 Granulomas in sarcoidosis express high levels of 1α-hydroxylase, an enzyme that catalyses
376 hydroxylation of 25-OH vitamin D to its active form, 1,25(OH)2 vitamin D [37], which has
377 been found positively associated with sarcoidosis [38]. VBDP is the primary transporter of
378 vitamin D and makes it available to specific tissues and cell types [39]. VDBP bound to
379 complement component 5a also has a chemotactic effect on neutrophils and macrophages [40,
380 41], and VDBP can act as a macrophage-activating factor (MAF) [42]. VDBP has been
381 previously detected in sarcoidosis patient BALF [26, 29] and plasma [43], although not in
382 connection with exosomes and not with any increase in the plasma. We found VDBP at two-
383 fold (by iTRAQ) higher levels in sarcoidosis patient BALF exosomes compared to healthy

384 individuals, and successfully validated these findings by ELISA and showed that VDBP was
385 not increased in any of the subjects in a control group of non-sarcoidosis lung patients (three
386 fibrosis patients, two with asthma and one with alveolitis). Diagnosing sarcoidosis is a
387 circumstantial task and better diagnostic procedures are warranted. To evaluate exosome-
388 associated VDBP as a biomarker in blood, we therefore investigated exosomes isolated from
389 sarcoidosis patient plasma, and found significantly higher levels of VDBP compared to in
390 healthy controls. The higher levels of VDBP in sarcoidosis plasma exosomes are not
391 attributed to increased numbers of exosomes, as shown by NTA, but rather enrichment for the
392 protein in the exosomes. The fact that only exosome-associated, but not total VDBP levels
393 were increased in patients further supports the use of exosomes as biomarkers, and if we will
394 be able to select organ-specific exosomes (i.e. from the lung) for analysis we might increase
395 specificity and sensitivity even further.

396

397

398 In conclusion, the observed differences in exosome protein expression between sarcoidosis
399 patients and healthy controls suggest a role for exosomes in several central inflammatory
400 systems, including leukotriene pathways. These findings may help to clarify the progress or
401 driving cause of sarcoidosis and provide new directions for its prophylaxis or treatment. We
402 also highlight exosome-associated complement components and VDBP as candidates for
403 sarcoidosis biomarkers. Diagnosing sarcoidosis is a complex task, and our finding that
404 exosomes may carry disease biomarkers even in the plasma of patients, opens up the
405 possibilities for both improved diagnostic and future studies on the implications of exosomes
406 in sarcoidosis.

407

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551

552

553 **FIGURE LEGENDS**

554

555 **Figure 1**: Protein profiles of BALF exosomes captured by anti-MHC class II coated latex
556 beads and analyzed by flow cytometry. **A-B,** Exosome-markers tetraspanin CD63 and CD9.
557 **C-D**, Expressions of HLA-DR and CD54 on exosomes. MFI= mean fluorescence intensity
558 normalized to isotype controls. Open circles are patients with Löfgren's disease, closed circles
559 repesent chronic sarcoidosis. *$p<0.05$, **$p<0.01$ by Mann-Whitney or Kruskal-Wallis test
560 (the latter for Löfgren's/non-Löfgren's/healthy individuals).

561

562 **Figure 2**: Summary of the biological processes affected in BALF exosomes from sarcoidosis
563 patients compared to healthy controls as measured by iTRAQ. DAVID, a functional
564 annotation clustering tool, was used to group relate biological processes. Annotation clusters
565 with an up and down-regulated expression score > 2 are shown.

566

567 **Figure 3**: Activating and inhibiting proteins involved in the complement cascade show a
568 different profile in BALF exosomes in sarcoidosis patients and healthy controls as measured
569 by ITRAQ. The abundance of the proteins is related to the internal control. Bars represent
570 relative linear protein abundance in sarcoidosis patients and healthy controls.

571

572 **Figure 4:** CD55 levels are lower in patient BALF exosomes than in healthy controls as
573 measured by iTRAQ and flow cytometry. **A**, Relative abundance of CD55 compared to a
574 pooled reference, measured by iTRAQ in sarcoidosis patients and healthy controls. **B**, CD55
575 levels on exosomes bound to anti-CD63-coated beads and measured by flow cytometry

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576 adjusted to isotype-matched control. Open circles are patients with Löfgren's disease, closed
577 circles represent chronic sarcoidosis. *$p<0.05$, **$p<0.01$ by Mann-Whitney or Kruskal-Wallis
578 test (the latter for Löfgren's/non-Löfgren's/healthy individuals).

579

580 **Figure 5:** Patient BALF exosomes carry more complement C3 compared to healthy controls.
581 **A**, Relative abundance of C3 compared to an internal pooled reference measured by iTRAQ
582 in sarcoidosis patients and healthy controls. **B**, C3 levels on exosomes bound to CD63-coated
583 beads and measured by flow cytometry adjusted to isotype-matched control. **C**, Western Blot
584 analysis of C3 in protein isolates from BALF exosomes of sarcoidosis patients (P1-P6) and
585 healthy controls (H1-H5), MW = molecular weight marker. Before transfer, the gel was
586 scanned for total protein contents, relative intensities correlating with protein amounts are
587 plotted in percent of the strongest sample. Open circles represent patients with Löfgren's
588 disease, closed circles are patients with chronic sarcoidosis. *$p<0.05$, **$p<0.01$ by Mann-
589 Whitney or Kruskal-Wallis test (the latter for Löfgren's/non-Löfgren's/healthy individuals).

590

591 **Figure 6:** BALF exosomes carry leukotriene pathway enzymes. **A**, LTA_4H in BALF
592 exosomes detected by iTRAQ in sarcoidosis patients and healthy individuals. Open circles
593 represent Löfgren's syndrome, closed circles represent patients with chronic sarcoidosis. **B**,
594 Western blot analysis of 5-LO and LTA4H in patients (S) and healthy controls (H). **C**,
595 Quantitative analysis of LTA_4H Western blot band intensities. *$p<0.05$, **$p<0.01$ by Mann-
596 Whitney or Kruskal-Wallis test (the latter for Löfgren's/non-Löfgren's/healthy individuals).

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598 **Figure 7:** Vitamin D binding protein (VDBP) is present on exosomes and is enriched in
599 patients compared to healthy controls. **A**, Relative abundance of VDBP on BALF exosomes
600 as detected by ITRAQ relative to a pooled reference. **B**, VDBP on BALF exosomes from
601 healthy controls and sarcoidosis patients as measured by ELISA. In the sarcoidosis group,
602 open circles represent patients with Löfgren's disease, and closed circles patients with chronic
603 disease. **C**, VDBP measured in plasma of patients and healthy controls by ELISA. **D**, VDBP
604 levels on exosomes isolated from plasma, as measured by ELISA. *$p<0.05$, **$p<0.01$ by
605 Mann-Whitney or Kruskal-Wallis test (the latter for Löfgren's/non-Löfgren's/healthy
606 individuals).

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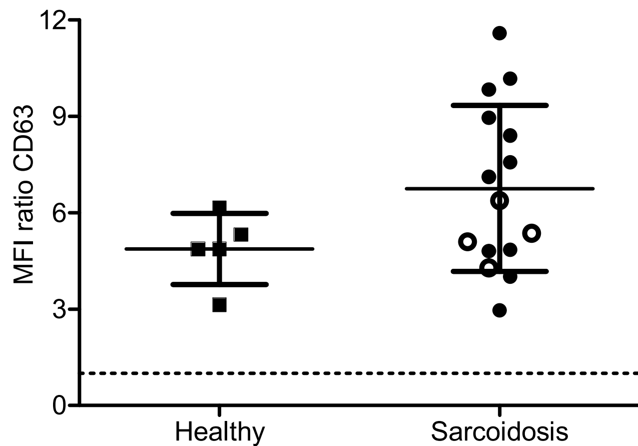

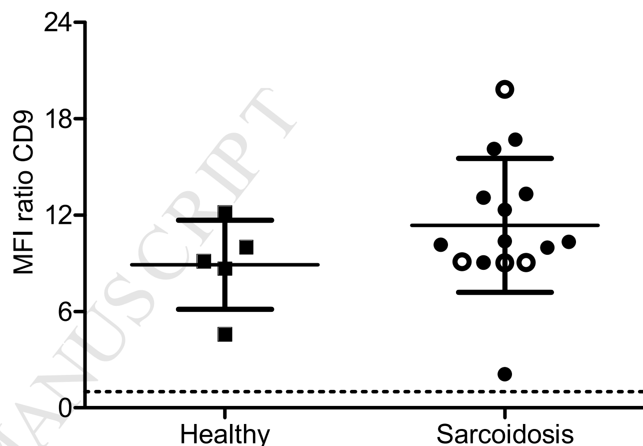

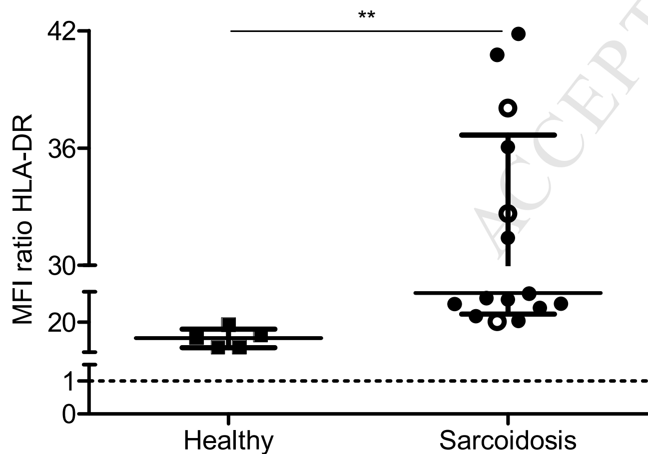

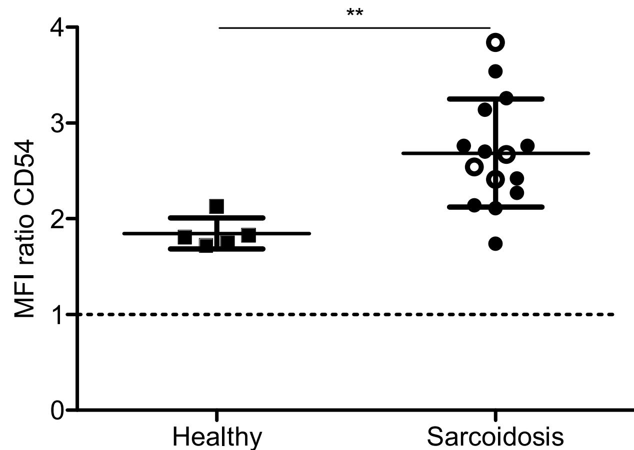

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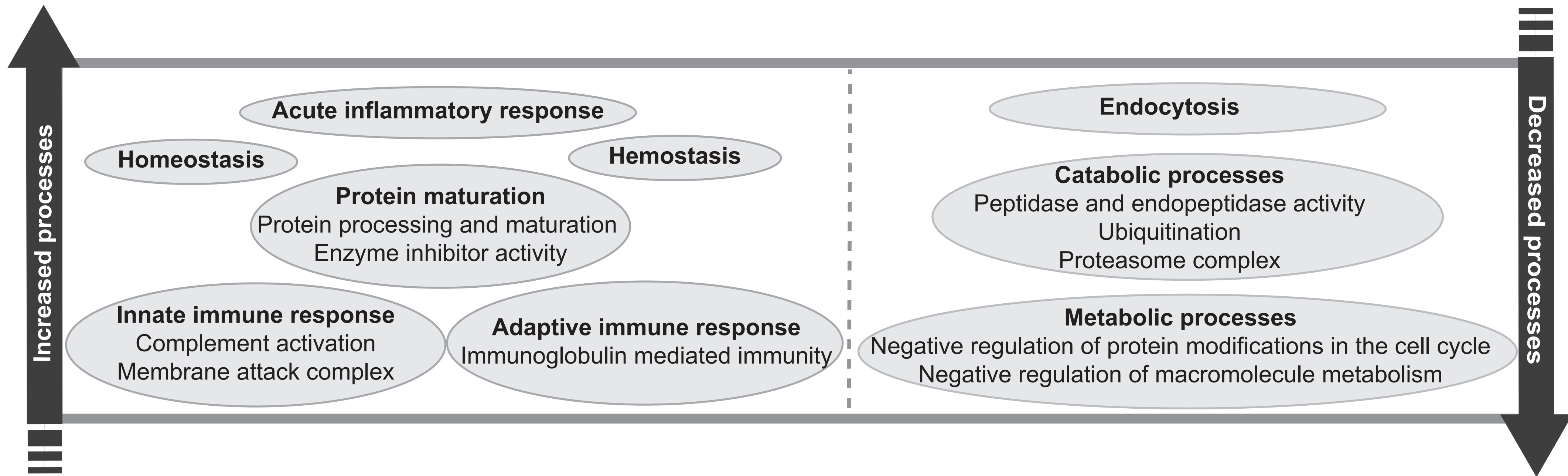

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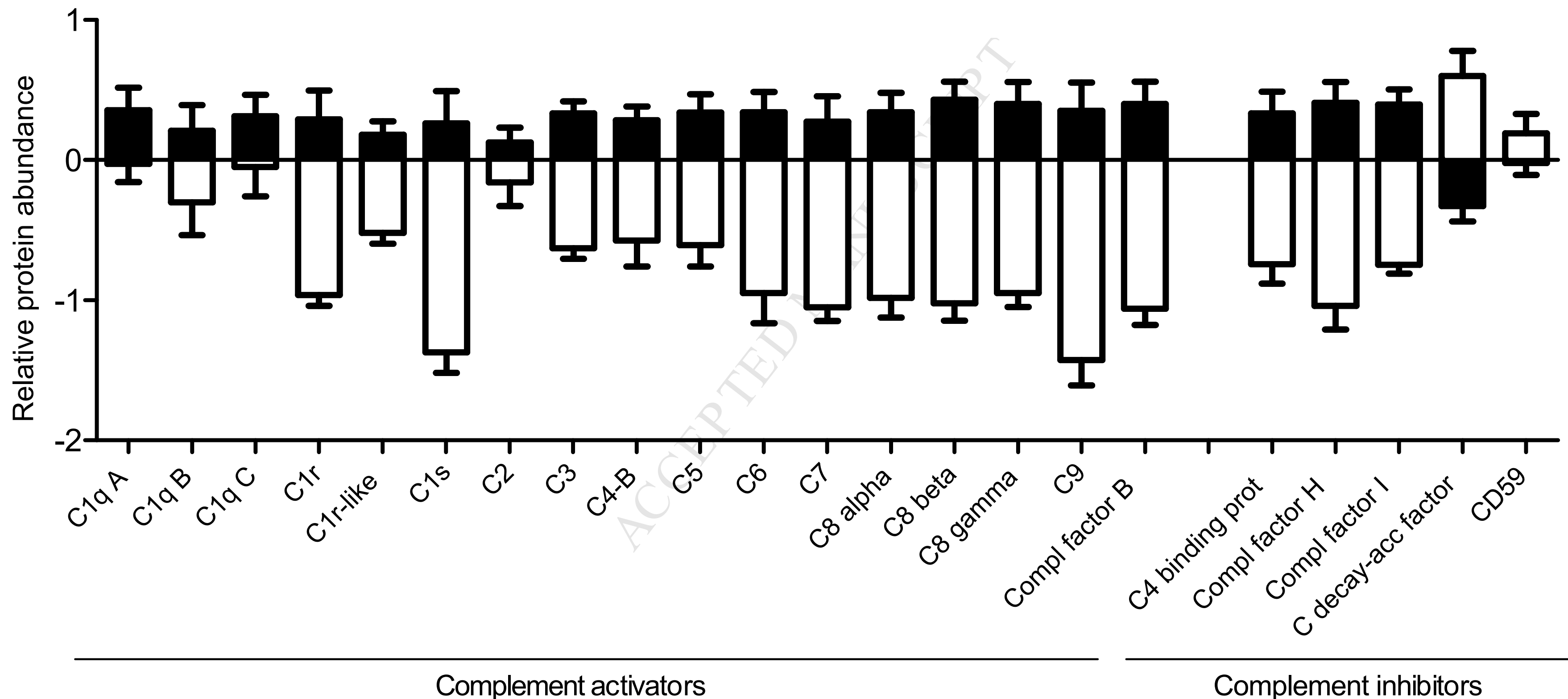

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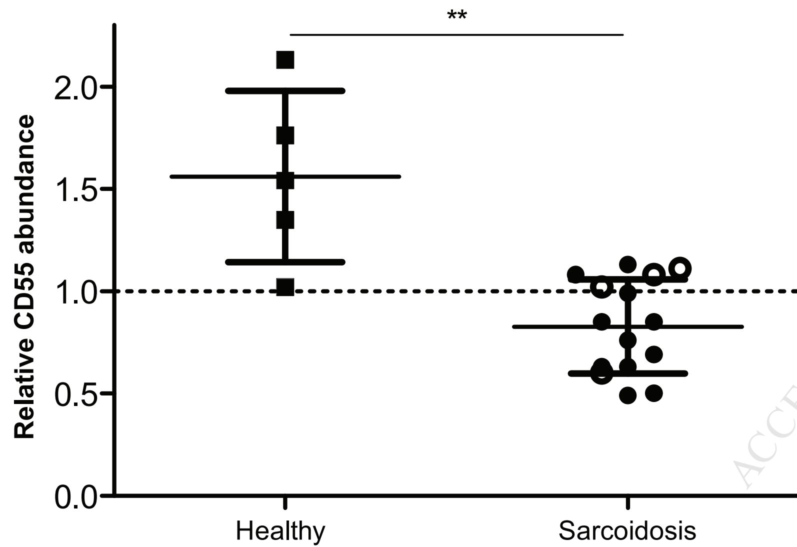

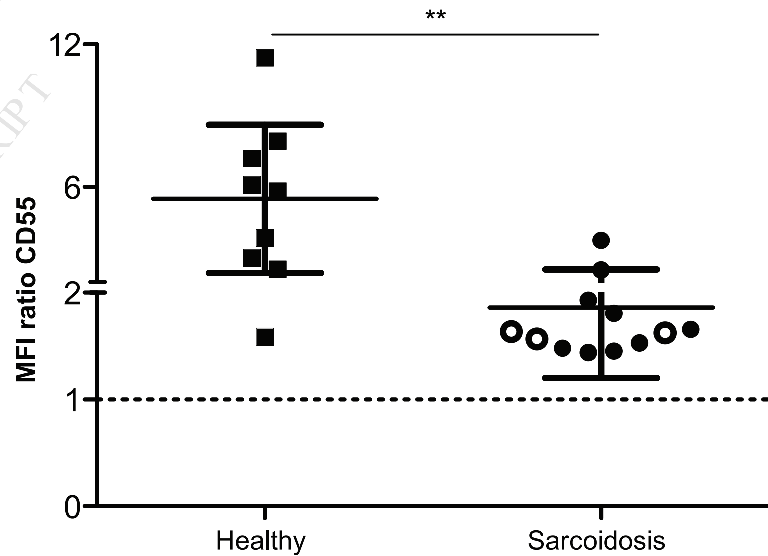

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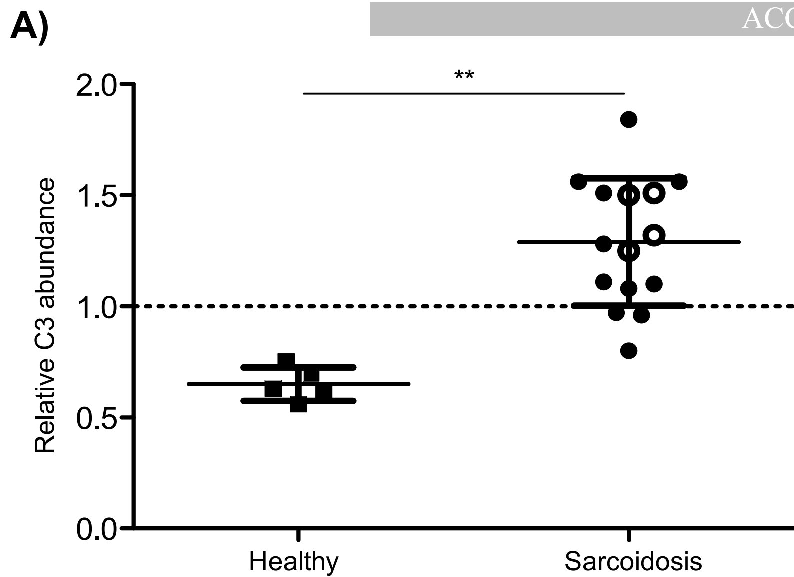
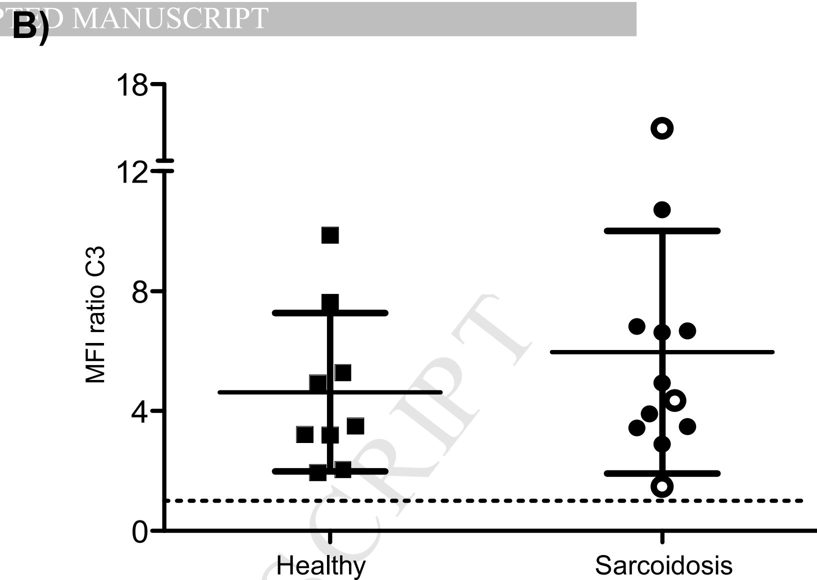
A)



B)



C)

MW H1 H2 H3 H4 H5 P1 P2 P3 P4 P5 P6

75 kD
50 kD
25 kD

62 64 40 33 18 43 21 38 45 100 75

Gel relative intensity %

250 kD
150 kD

C3, 187 kD

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A)

Relative LTA4H abundance

2.0
1.5
1.0
0.5
0.0

*

Healthy
Sarcoidosis

B)

S H S H S H S S H S H S

LTA4H
5-LO
β-actin
FLAP

C)

LTA4H band intensity

0.8
0.6
0.4
0.2
0.0

*

Healthy
Sarcoidosis

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A)

**
2.0
1.5
1.0
0.5
0.0
Relative VDBP abundance
Healthy
Sarcoidosis

B)

**
3000
2000
1000
0
VDBP /ng(ml)
Healthy
Sarcoidosis

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C)

*
1000
800
600
400
200
0
VDBP, mg/ml
Healthy plasma
Sarcoidos plasma

D)

**
100
80
60
40
20
0
VDBP /ng(ml)
Healthy plasma exo
Sarcoidosis plasma exo

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Detailed methods

Sample preparation and iTRAQ labeling

The BALF exosomes (50 µg per subject) from 15 sarcoidosis patients and 5 healthy controls were precipitated using acetone (6:1), dissolved in a denaturing buffer (6 M urea, 0.05% SDS, 5 mM EDTA, and 50 mM Tris-HCl (pH 8.5)), and reduced with 1 mM Tris 2-carboxyethyl phosphine hydrochloride for 2 hours minutes at 37°C, followed by alkylation with the addition of 1 µl of 200 mM MMTS and incubation for 10 min at room temperature. Proteins were digested overnight (16-h) with trypsin (20:1, sequencing grade modified trypsin) (Promega, Madison, WI, USA) at 37°C. The digests were purified using Sep-Pak® Vac C18 cartridges (1cc/50mg) (Waters, Milford, MA, USA), dried and re-dissolved in 0.5 M TEAB (30 µl). The purified digests were assayed at 280 nm with a Nanodrop detector (Thermo Scientific, Waltham, MA, USA) and the concentrations adjusted such that equal (estimated) amounts of the peptides would be labelled (40 µg). A sub-aliquot of each unlabelled digest was taken to create a reference pool that could be used to cross compare samples across separate iTRAQ experiments. The labeling was performed and mixtures composed such that the 15 cases and 5 controls were divided between three 8-plex experiments, each with one reference pool (113 reporter), two controls (or one control and one control pool) and five cases.

Small aliquots of the labelled samples were analyzed by LC-MS/MS to confirm efficacy of the labelling experiments before proceeding. The labelled samples were mixed (as indicated in below, Supplementary Figure 6), dried and acidified. These mixtures were fractionated by strong cation exchange chromatography using a 200 mm x 4.6 mm i.d. PolySULFOETHYL-A column with a BioCAD HPLC system, as previously described (1). Forty SCX fractions were collected, dried and desalted using OMIX C18 100-µl tips and pooled on the basis of UV absorption at 280 nm, determined with a Nanodrop detector (Thermo Scientific, Waltham, MA, USA).

LC-MS/MS Analysis

i) A QSTAR-Elite was used together with an Ultimate 3,000 capillary LC was used for LC-MS/MS. Spectra were recorded between 350 to 1,400 m/z for the top five most intense fragment ions with the enhance iTRAQ setting applied.

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A 150 mm x 75 µm i.d. tapered column packed with Magic C18 (200Å) was used in combination with a 2 mm x 300 µm C18 pre-column (LC-Packings, Amsterdam, Netherlands). The separation buffers were 98% water, 2% acetonitrile and 0.2% formic acid for the A-phase, and 95% acetonitrile, 5 % water with 0.2% formic acid for the B-phase. The separation gradient was from 2 to 35% B in 68 minutes at a flow rate of 200 nl/min, (further details are indicated in the supplementary information). The QSTAR was used to confirm labelling efficiency and for preliminary analyses of the labelled samples.

ii) An Orbitrap-Velos coupled with an EasyNano-LC was also used for LC-MS/MS. The MS/MS data were acquired in positive ion mode with a data dependent acquisition setting for HCD of the 10 most intense ions (m/z 300-2,000) with charge states of 2+ or higher. MS1 spectra were acquired with the Orbitrap set to a resolution of 30,000 (at m/z 400), with a target value of 1,000,000 ions and a maximal injection time of 100 ms. MS/MS spectra were acquired in the Orbitrap with a resolution of 7,500 (at m/z 400), a target value of 50,000 ions, a maximal injection time of 200 ms, and the lowest mass fixed at m/z 100. Dynamic exclusion was set to 60 s.

A 75 µm x 150 mm analytical column packed with 5 µm Magic C_{18} (Michrom, Auburn, CA, USA), coupled together with a 20 x 0.1 mm i.d. pre-column packed with 5 µm Magic C_{18} (Michrom, Auburn, CA, USA) silica, was used with a gradient from 5 to 40% B in 100 min, at a flow rate of 300 µl/min. The Orbitrap was used for analysis of the SCX fractions and thus produce the data used in the final quantitative comparisons.

LC-MS/MS Data Analysis

The QSTAR data were directly analysed using ProteinPilot™ software and the Paragon™ identification algorithm (2) with a human Swiss-Prot database (Release August 18th 2011, 20245 entries including 162 common non-human contaminants). The database searches were made in thorough mode specifying 8-plex-iTRAQ quantification, trypsin digestion and MMTS modification of cysteine. The Orbitrap data were analysed with Proteome Discover (version 1.3) together with Mascot (Matrix Science, Boston, MA, USA) specifying 8-plex-iTRAQ quantification, trypsin digestion and MMTS modification of cysteine, deamidation of N/Q and methionine oxidation, using the same database. The mass tolerance settings of 5 ppm for the precursors and 0.02 Da for fragments were used. To accommodate analysis of the Orbitrap data with ProteinPilot, the files were converted to MGF format using Proteome Discover version 1.3 (Thermo Scientific, Waltham, MA, USA) and filtered using the script PeakConvert, as described elsewhere (3). False discovery rates for identification were calculated with the ProteinPilot™ PSEP functionality, based on the relative proportion of identification from the forward and reverse sequence searches (4). A confidence threshold of 95% for protein identification was applied.

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ITRAQ ratios were calculated relative to the pooled reference using both ProteinPilot™. ProteinPilot™ facilitated the combination and co-analysis of data from the different mass spectrometers and was thus chosen. The ProteinPilot™ ratio calculations are based on the weighted average ratio of the contributing peptides, as previously described (1, 5). The calculations excluded values that were blanks, zeros, greater than 99, or for which the sum of the peak areas is less than 40 counts. Only peptides sequences that were unique were used for protein quantification. Peptides that indicated non-tryptic cleavage, or contained a low probability modification were also removed. A cut off excluding peptides identified with less than 65% confidence from quantification was applied. Data normalization was made to correct for possible systematic bias. The software package R (http://www.rproject.org/) was used to merge the data. The ProteinPilot™ Descriptive Statistics (6) and Protein Alignment Templates were used to asses labelling efficiency and compare iTRAQ experiments, respectively. The mass spectrometry proteomics data have been deposited to the ProteomeXchange Consortium (7) via the PRIDE partner repository with the dataset identifier PXD001808 (http://www.ebi.ac.uk/pride/archive/). The data was converted using PRIDE Converter2 (8, 9) (http://pride-converter.googlecode.com).

SDS-PAGE and western blot for leukotriene forming enzymes

Exosome aliquots were mixed with 4x Laemmli buffer and heated for 5 min at 95°C. Typically, 7.5 µg of exosomal proteins were loaded for each sample and separated on SDS-PAGE 4-20% pre-cast gels (BioRad Laboratories, Hercules, CA, USA). After electroblotting, the nitrocellulose membranes (GE Healthcare, Amersham Place, Buckinghamshire, UK) were blocked for 1 h with 5% dry milk powder in Tris buffered saline with 0.1% Tween-20. The following antibodies were used: in house antisera (rabbit polyclonal) against 5-LO (1:300 dilution), FLAP (1:300), LTA_4H (1:750), peroxidase-conjugated primary antibody against β-actin (1:2000 dilution, Sigma-Aldrich, St Louis, MO, USA) and IRDye® 800CW antirabbit secondary antibody (1:10000) (Li-COR, Lincoln, NE, USA). Protein bands were detected by the LI-COR Odyssey IR Imaging System.

Western blot for complement component C3

Exosome aliquots were mixed with RIPA buffer, sonicated and vortexed in cycles to isolate exosomal proteins. Four micrograms of total proteins were mixed in 4X non-reducing Laemmli buffer and separated on a pre-cast any kD gel (BioRad Laboratories, Hercules, CA, USA) at 240 volts for 30 minutes, and total protein contents of the gel was visualized using a stain-free scan technology (BioRad Laboratories, Hercules, CA, USA). The proteins were then transferred to a PVDF membrane in a semi-dry Turblot system (BioRad Laboratories, Hercules, CA, USA). The membrane was blocked and incubated with primary mouse anti-human C3 (Abcam Ltd). As a control for non-specific binding, an identical second gel was run in parallel and treated identically but without primary antibody. Secondary ECL sheep anti-mouse HRP-conjugated antibody (GE Healthcare, Amersham Place, Buckinghamshire, UK) and an ECL developing kit

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(BioRad Laboratories, Hercules, CA, USA) were used before scanning the membranes using a ChemiDoc scanner (BioRad Laboratories, Hercules, CA, USA).

Nanoparticle tracking analysis

An LM10 instrument equipped with an LM14 laser unit (Nanosight/Malvern, UK) was used to record vesicle samples under constant flow induced by a syringe pump. The NTA 3.0 software was used to analyse the videos and calculate vesicle size distributions and concentrations. Samples were diluted to vesicle concentrations between $(1\text{-}10)^8$ vesicles/ml and 30-50 particles per frame, and five 60 second videos were recorded per sample at camera level 13. Batch processing was used to analyse the videos at threshold level 5, resulting histograms were overlaid and the average vesicle concentration of each batch was used to compare relative vesicle quantities between patients and controls.

Bioinformatics

The Reproducibility-Optimized Test Statistic (ROTS) was used for these analyses (10) to determine protein abundances between the healthy subjects and sarcoidosis patients. With this method the test statistic is interpolated from the inherent distribution of the data and does not assume normal distribution of the data. Comparisons were made between exosome samples form healthy subjects versus those from Löfgrens-Sarcoidosis patients and from non-Löfgrens Sarcoidosis patients. The Löfgrens-Sarcoidosis and non-Löfgrens samples were also compared. The Orbitrap data was used in the quantitative comparisons.

GO annotation and Pathway analysis: Ingenuity pathway analysis and Database for Annotation, Visualization and Integrated Discovery (DAVID) (http://david.abcc.ncifcrf.gov/) were used to identify common pathways and functional annotation associated with the protein identified as differentially abundant between the Sarcoidosis subjects and controls (ROTS analysis, false discovery rate of <0.05).

To identify differences between two groups, the non-parametric Mann-Whitney test was used. For comparing three groups the Kruskal-Wallis test was performed using GraphPad Prism 5 (GraphPad Software, Inc. La Jolla, CA, USA). P<0.05 was considered significant.

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Table E1. Demographic and clinical data of subjects included in the study

BALF exosomes from patients 1 to 15 and healthy 1 to 5 (healthy subjects on next page) were analyzed by iTRAQ. Patients 48-53 served as non-sarcoidosis lung disorder controls for plasma exosome VDBP levels. Smoking status: no, never has smoked; ex, stopped smoking more than 5 years ago; yes, current smokers. Missing data are represented by -. Normal Ca^{2+} levels in blood are 2.2-2.6 mmol/L. Normal Ca^{2+} ion free values are 1.11-1.30 mmol/L. Disease stages are confirmed by radiology findings.

											BAL cells							Spirometry					
Patient	Age	Sex	Smoker	CD4/CD8 Ratio	Ca2+ in blood	Ca2+ free	S-ACE kE/L	Löfgrens?	Stage	10^6 cells per L	Macroph	Lympho	Neutro	Eosino	Baso	Mast	VA23 %	VC%	FVC%	TLC%	FEV1%	DLCO%	FEV1/VC
P1	41	**F**	Yes	4.1	2.45	1.29	131.3	No	2	278,6	81	18,2	0,8	0	0	3	17	56	55	71	51	58	76
P2	45	**F**	Ex	14	2.35	1.26	57.8	Yes	1	186,2	78	21	1	0	0	0	31	80	90	-	79	-	83
P3	45	**F**	Yes	7.3	2.31	1.27	81.6	No	1	170,6	79,4	20,4	0	0	0	0	7,3	110	11	122	107	85	78
P4	51	**F**	Ex	6.5	2.34	1.28	90.3	Yes	2	210	59	32,6	1,2	6,8	0,4	56	13	79	78	91	72	65	75
P5	69	**F**	Ex	5.8	-	-	-	No	-	185	65	32,8	0,4	1,2	0,6	10	5,2	-	-	-	106	-	76
P6	29	M	Ex	12	2.28	-	224.2	No	-	210,4	55,4	44,2	0,4	0	0	3	2,9	-	10	-	103	-	79
P7	37	M	Ex	9.5	-	-	-	No	-	202,2	41	55,4	3,2	0,4	0	0	3,6	59	-	73	31	64	41
P8	38	M	N	8	2.46	1.3	-	Yes	2	279	42,6	55,4	1,2	0,4	0,4	5	2,6	100	97	-	96	-	73
P9	39	M	N	3.3	2.24	1.25	81.8	No	2	136,3	77,2	20,8	1,6	0,4	0	0	4,2	89	10	-	103	-	90
P10	41	M	Yes	14	2.38	1.27	96.9	Yes	1	323	76	22	0,6	1,4	0	12	2,2	88	-	88	94	107	81
P11	44	M	N	10	2.34	1.25	163.1	No	2	264	55	43,6	0,9	0,5	0	0	1,2	-	-	-	-	-	-
P12	51	M	N	21	2.49	1.36	85.3	No	2	205	66	29,6	2,3	2,1	0	11	1,8	-	92	-	96	-	-
P13	57	M	N	13	2.27	1.24	143.1	No	1	168,2	46	50,5	1,3	1,7	0,2	0	3,5	-	73		63		69
P14	65	M	N	4.7	2.27	1.24	87.6	No	2	236,3	58,6	19	19,5	2,9	0	5	9,8	75	77	77	72	76	70
P15	69	M	N	10	2.26	1.21	61.2	No	1	110	56,2	41,8	1,6	0,4	0	0	4	64	10	-	69	-	80
P16	26	F	Ex	7,7	2,36	-	77,9	No	3	137,9	73,6	24,6	0,6	1,2	0	0	4,6	93	-	94	100	97	90
P17	31	F	Ex	5,7	-	-	-	Yes	2	306,7	79,2	20,2	0,4	0,2	0	2	24	106	-	100	104	75	79
P18	34	F	No	-	-	-	-	No	-														
P19	35	F	No	4,8	-	-		Yes	2	100	84,6	13,2	1,4	0,8	0	0	7,5	-	-	-	-	-	-
P20	36	**F**	No	5.1	2.24	1.22	57.2	No	0	88,3	82	16,4	1,6	0	0	0	3,6	89		108	83	97	
P21	36	F	Yes	9,1	-	1,26	84,3	Yes	1	171,6	89,4	10,2	0	0,2	0	2	34,4		13		119		79
P22	37	F	No	15.6	2.24	1.27	-	No	1	98	64,3	25,3	10	0,4	0	0	15,6	74	78		81		91
P23	38	F	No	18	-	-	-	Yes	1	388	59,7	38	2,1	0,2	0	0	2,7	72	71	68	64	66	
P24	39	F	No	13,9	2,34	-	-	No	1	104,4	75,6	23,2	0	1,2	0	0	2,5	77	83		85		84

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P25	40	F	No	17	2.26	1.26	-	Yes	2	197	51,2	46	1,2	1,6	0	2	2,9	57	58	90	73	84	99
P26	45	F	Ex	14,7	2,23	-	95,3	Yes	1	225,8	59,7	39,5	0,8	0	0	1	21	77	75		61		60
P27	48	F	No	8.3	2.35	-	-	No	3	179	91	8,2	0,2	0,6	0	3	9,2	76	74	83	88	82	93
P28	50	F	No	12.1	-	-	-	No	2	126,5	76,4	20,4	1,8	1,4	0	0	2,5	94	93		78		64
P29	52	F	No	18.7	-	1.28	105.7	Yes	2	416	53,6	40,8	4,6	0,8	0,2	1	33		85		88		
P30	56	F	No	9.2	-	1.31	126.5	Yes	2	145,6	57,7	41,5	0,8	0	0	0	14						
P31	59	F	No	9,7	-	-	-	No	2	192	57,7	42	0	0,3	0	0	4	84		92	82	70	77
P32	78	F	No	4.9	2.23	-	35.4	No	2	275,7	56,6	42,6	0,3	0,3	0,2	0	7,1						
P33	28	M	Ex	8,5	-	-	-	No	2	101,8	80	18,3	1,3	0	0	0	3	93	11		117		79
P34	29	M	Ex	26.4	2.32	1.26	-	Yes	1	152,2	62,7	36,5	0,5	0,3	0	3	39	94	92	91	90	102	75
P35	31	M	No	3	2.53	1.21	76.6	No	2	156,7	93,8	5,4	0,8	0	0	4	14,9						
P36	33	M	No	9.2	2.36	1.33	76	No	2	332,6	44	53,7	1,3	1	0	0	14	92		84	83	84	72
P37	41	M	Yes	6	2.32	-	-	No	2	693	75,6	23,4	0,8	0,2	0	39	13	84		84	85	95	76
P38	43	M	Ex	14,5	2,36	-	137	No	2	125	78,6	20,4	0,2	0,8	-	7	3,2	71	60		69		74
P39	44	M	No	3.2	2.38	-	77.1	No	2	437,6	65,2	34,5	0,3	0	0	0	1,4	90	90		87		74
P40	45	M	No	15.2	2.17	-	-	Yes	1	272,3	68,5	28	3,5	0	0	1	35,3						
P41	46	M	No	6.3	2.23	1.24	-	Yes	1	165,7	80,5	17	2,5	0	0	5	26	67	64	86	63	91	71
P42	46	M	No	1,5	2,32	-	34,7	No	1	323	76	22	0,6	1,4	0	12	2,2	88		88	94	107	81
P43	46	M	Yes	0,8	2,27	-	54	No	2	384,6	89,8	9,4	0,8	0	0	0	3,3						
P44	48	M	Ex	7.8	2.46	1.3	-	No	1	252,7	81,6	16,8	1,2	0,2	0,2	4	3,1	74	77		84		85
P45	**49**	M	No	8,6	2,38	-	-	Yes	1	123	70,4	21	0,8	7,6	0,2	11	8,5				92		
P46	52	M	No	9.6	2.63	1.5	-	No		620,2	78,2	20,2	1,6	0	0	30	4,5	62		65	66	60	80
P47	68	M	Ex	37,1	2,28	-	72,7	No	2	231,4	59,6	39,2	1	0	0,2	0	2,3		66		65		
N1	20	F	No	-	-	-	-	Asth		44,2	86,4	10,7	2,7	0,2	0	8	-	102	10		82		
N2	25	M	No	-	-	-	-	Asth		-	83,5	3,5	13	0	0	0	-	92	10		94		
N3	69	F	Ex	2.1	2.36	-	-	Fibro	-	335	75	22,8	0,6	1,4	0,2	19	3	69	66		65		72
N4	75	M	Ex	1.5	2.34	-	-	Fibro	-	427,1	72,2	20,6	5	1	1,2	63	-	105	10	90	109	78	74
N5	50	F	No	17,6	2,29	-	-	Alve		163,4	56	33,2	10,6	0,2	0	13	-	67	63	80	45	74	54
N6	72	F	No	3,2	2,41	-	-	Fibro		464	49,6	39,8	5,2	4	1,4	16	6,1	77	78		78		

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Table E1 continued, healthy controls.

						BAL cells								Spirometry	
Healthy	Age	Sex	Smoker?	CD4/CD8	Ca2+ in blood	10^6 cells per L	Macroph	Lympho	Neutro	Eosino	Baso	Mast	VA23 %	FVC%	FEV1 %
H1	23	F	N	2		52,1	92,6	6	1,4	0	0	3	-	104	100
H2	24	F	N	2.7	-	67,4	95	3,2	1,8	0	0	0	-	98	103
H3	23	M	N	1.6	-	39,7	97	2,3	0,7	0	0	0	-	108	107
H4	25	M	N	-	-	110	77	8	14,8	0	0,2	1	-	113	91
H5	30	M	N	1.2	-	81,3	89,6	9,8	0,6	-	-	1	-	106	109
H6	20	F	No	1,4	-	83,1	73,6	25	1,4	0	0	0	2,8	105	97
H7	21	F	No	2.9	2.39	111,1	91	7,6	1	0,4	0	0	-	123	126
H8	24	F	No	3.2	2.4	44,8	97,8	2	0,2	0	0	0	-	112	111
H9	25	F	N	2,6	-	204,5	91,3	2,6	5,5	0,6	0	0	-	102	101
H10	39	F	No	2.9	2.35	56,4	86	10,3	3,7	0	0	0	-	92	95
H11	42	F	No	3,7	-	105,8	89,3	5,7	5	0	0	0	1,3	115	119
H12	52	F	N	2,5	-	89,2	84	14,5	1,5	0	0	0	-	91	84
H13	18	M	No	1.4	2.39	62	88,3	10,6	1,1	0	0	0	4,5	118	107
H14	20	M	N	1,1	-	57,4	95,3	3,6	0,8	0,3	0	2	-	106	89
H15	21	M	N	3,3	-	68	89	9,6	1,4	0	0	2		88	103
H16	22	M	No	0.7	2.32	76,8	93	4,8	2	0	2	0	2,9	115	113
H17	22	M	Yes	2.4	2.35	61,9	90,6	8,8	0,4	2	0	5	3,9	106	106
H18	22	M	No	2,6	-	86,98	88,3	11,3	0,4	0	0	6	-	130	125
H19	23	M	No	7	2.48	95,4	85,6	13,3	1,1	0	0	0	-	122	117
H20	24	M	N	4,8	-	85,4	87,1	11,3	0,6	1	0	0		128	103
H21	24	M	No	1,4	-	72,3	89,3	10	0,7	0	0	0	-	108	108
H22	24	M	No	2.4	2.35	79,8	87,3	11,6	0,9	0	0,2	7	-	85	73
H23	26	M	N	4,2	-	152,5	66,6	31,6	1,8	0	0	0		86	89
H24	26	M	N	1,3	-	61,7	93,6	6,3	0,1	0	0	1		107	109
H25	29	M	No	1.5	2.32	39,5	86,2	12,6	0,3	0,9	0	0	-	121	112
H26	52	M	Yes	-	2.33	570,7	95,6	4,2	0,2	0	0	2	-	122	122
H27	53	M	Yes	0,7		910,8	93,2	5,4	1,2	0,2	0	4	-	99	80

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Table E2a. List of proteins two-fold or more increased in abundancy in 15 sarcoidosis patients compared to in 5 healthy individuals.

ID	Protein Name	FDR	Average fold difference	Protein Score	% Sequence coverage	Unique peptides	Average Calculations
P02748	Complement component C9	0	3.01	22.91	24.0	15	31
P02760	Protein AMBP	0	2.95	15.74	28.7	9	29
P01042	Kininogen-1	0	2.88	19.7	21.1	15	19
P05546	Heparin cofactor 2	0	2.86	34.04	42.5	24	36
P09871	Complement C1s subcomponent	0	2.85	8.7	13.4	6	6
P02751	Fibronectin (FN) (Cold-insoluble globulin) (CIG)	0	2.79	175.92	49.4	191	287
P18428	Lipopolysaccharide-binding protein (LBP)	0	2.73	11.18	16.0	7	16
Q96PD5	N-acetylmuramoyl-L-alanine amidase	0	2.64	18.55	31.8	11	16
Q15063	Periostin (PN)	0	2.64	35.61	32.5	28	41
P19827	Inter-alpha-trypsin inhibitor heavy chain H1	0	2.62	34.36	24.2	23	37
P02765	Alpha-2-HS-glycoprotein	0	2.61	21.95	43.9	16	31
P27169	Serum paraoxonase/arylesterase 1	0	2.60	11.89	25.9	6	14
P01023	Alpha-2-macroglobulin	0	2.52	270.35	74.6	416	898
P01011	Alpha-1-antichymotrypsin	0	2.51	30.79	47.7	30	51
P00751	Complement factor B	0	2.50	47.72	38.1	34	49
P19823	Inter-alpha-trypsin inhibitor heavy chain H2	0	2.49	39.17	23.1	30	35
P00734	Prothrombin	0	2.48	27.9	31.3	17	22
P08697	Alpha-2-antiplasmin	0	2.48	6.28	12.2	4	8
P02763	Alpha-1-acid glycoprotein 1	0	2.47	19.67	44.8	18	45
P07358	Complement component C8 beta chain	0	2.46	27.73	27.8	15	46
P06276	Cholinesterase	0	2.42	6.47	7.1	3	8
P02749	Beta-2-glycoprotein 1	0	2.40	16.88	29.6	13	17
P04196	Histidine-rich glycoprotein	0	2.40	25.97	31.2	18	38
P01860	Ig gamma-3 chain C region	0	2.34	14.45	72.9	140	23
Q14624	Inter-alpha-trypsin inhibitor heavy chain H4	0	2.32	34.03	25.8	20	21
P08603	Complement factor H	0	2.32	81.34	41.7	53	54
P11678	Eosinophil peroxidase	0	2.30	20.83	20.0	17	18
P00736	Complement C1r subcomponent	0	2.30	6.7	6.7	5	6
P01857	Ig gamma-1 chain C region	0	2.29	97.27	86.7	211	257
P02647	Apolipoprotein A-I	0	2.28	36.95	63.7	39	75
P02652	Apolipoprotein A-II	0	2.27	6.24	37.0	7	36
P00748	Coagulation factor XII	0	2.25	13.06	17.1	8	7
P10643	Complement component C7	0	2.25	29.5	28.2	21	25
P00747	Plasminogen	0	2.24	56.98	43.3	34	41
P07360	Complement component C8 gamma chain	0	2.24	9.88	49.0	7	8
P02679	Fibrinogen gamma chain	0	2.23	38.41	49.5	33	63
P13671	Complement component C6	0	2.23	9.1	9.0	7	6
P19652	Alpha-1-acid glycoprotein 2	0	2.22	9.7	43.8	11	10

P07357	Complement component C8 alpha chain	0	2.22	18.77	22.9	12	11
P01008	Antithrombin-III	0	2.21	39.03	46.3	27	49
P01009	Alpha-1-antitrypsin	0	2.21	71.15	69.4	89	184
P00450	Ceruloplasmin	0	2.20	74.74	46.8	65	150
P02671	Fibrinogen alpha chain	0	2.20	48.6	29.1	53	67
P00738	Haptoglobin	0	2.19	72.73	72.9	98	183
P55058	Phospholipid transfer protein	0	2.16	17.79	16.8	10	20
P36955	Pigment epithelium-derived factor	0	2.15	18.54	30.9	13	19
P43652	Afamin	0	2.08	25.67	29.2	15	22
P29622	Kallistatin	0,02	2.07	13.62	23.2	8	13
Q8N4F0	BPI fold-containing family B member 2	0,02	2.06	4.12	12.0	3	5
P01859	Ig gamma-2 chain C region	0	2.03	36.41	84.0	161	52
P02774	Vitamin D-binding protein	0,02	2.00	54.5	63.1	41	89

Table E2b. List of proteins two-fold or more decreased in abundancy in 15 sarcoidosis patients compared to in 5 healthy individuals.

ID	Protein Name	FDR	Average fold difference	UnUsed Protein Score	% Sequence coverage	Unique peptides	Average Calculations
O75531	Barrier-to-autointegration factor	0	4.42	8.68	65.2	8	12
P49721	Proteasome subunit beta type-2	0	2.35	11.67	41.3	10	11
Q53TN4	Cytochrome b reductase 1	0	2.15	6.96	18.5	11	8
P28070	Proteasome subunit beta type-4	0	2.09	8.1	21.2	5	9
Q03135	Caveolin-1	0	2.08	9.79	40.5	7	7
Q8TDB8	Solute carrier family 2	0	2.01	4.47	3.5	2	4
P20618	Proteasome subunit beta type-1	0.0149	2.01	16.95	38.6	13	30

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Table E3: ITRAQ Labelling scheme used to compare BALF exosome samples.

Experiment	iTRAQ Reporter ion							
iTRAQ 1	113	114	115	116	117	118	119	121
	pool	Control 1	Control 2	Sar.1	Sar.L1	Sar.2	Sar.3	Sar.L2
iTRAQ 2	113	114	115	116	117	118	119	121
	pool	Control 3	Control 4	Sar.4	Sar.5	Sar.6	Sar.L3	Sar.7
ITRAQ 3	113	114	115	116	117	118	119	121
	pool	Control 5	Sar.8	Sar.9	Sar.10	Sar.L4	pool.2	Sar.11
Sar.L1, Sar.L2, Sar.L3 and Sar.L4 are Löfgrens Sarcoidosis samples Control 1, 2, 3, 4 & 5 = healthy subjects. Sar.1 to 11 = non-Löfgrens Sarcoidosis								

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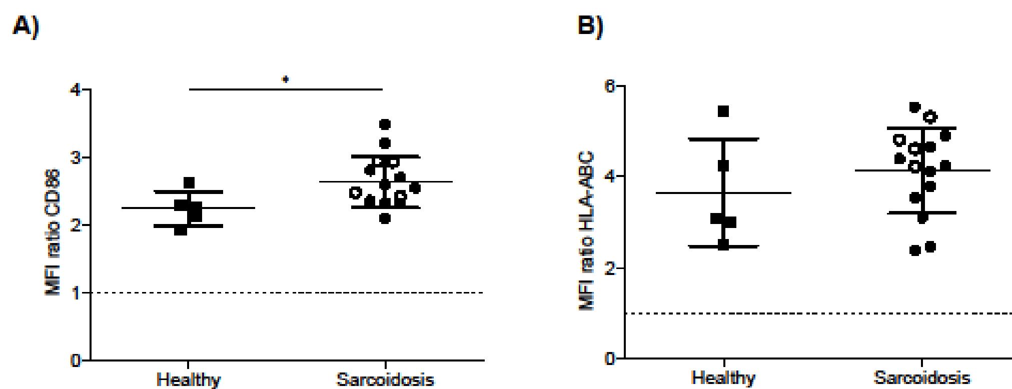
Figure E1. Surface expression of CD86 and HLA-ABC on BALF exosomes.



The BALF exosomes included in the iTRAQ analysis were analyzed by flow cytometry for CD86 (A) and HLA-ABC (B), showing presence of both markers on exosomes from healthy and patients, with a slightly higher level of CD86 on patient exosomes. Statistical analysis was performed by a Mann-Whitney test, * $p<0.05$

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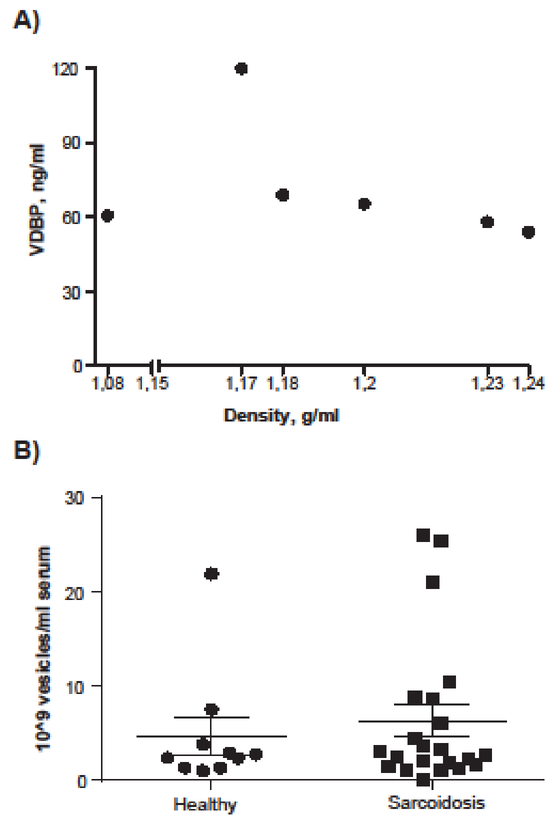
Figure E2. VDBP is associated to exosomes, and patients do not have more vesicles in plasma than healthy controls.



(A) A pool of exosomes from five patients was separated on a continuous gradient of sucrose, fractions corresponding to exosome densities were washed and investigated for VDBP levels, showing VDBP in exosome-associated densities. (B) Plasma exosomes from healthy controls and patients were analyzed by nanoparticle tracking analysis, showing no significant difference between the groups.

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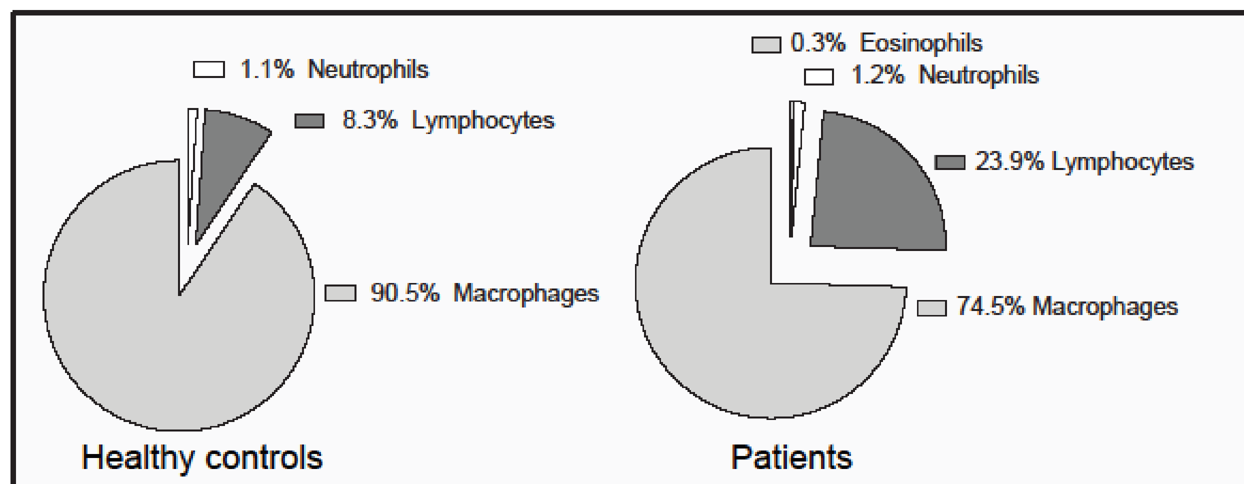
Figure E3. BAL cell type proportions of healthy controls and patients.



The cells from the BAL fluid collected from the patients and healthy controls listed above in supplementary table 1 analyzed by flow cytometry and summarized.

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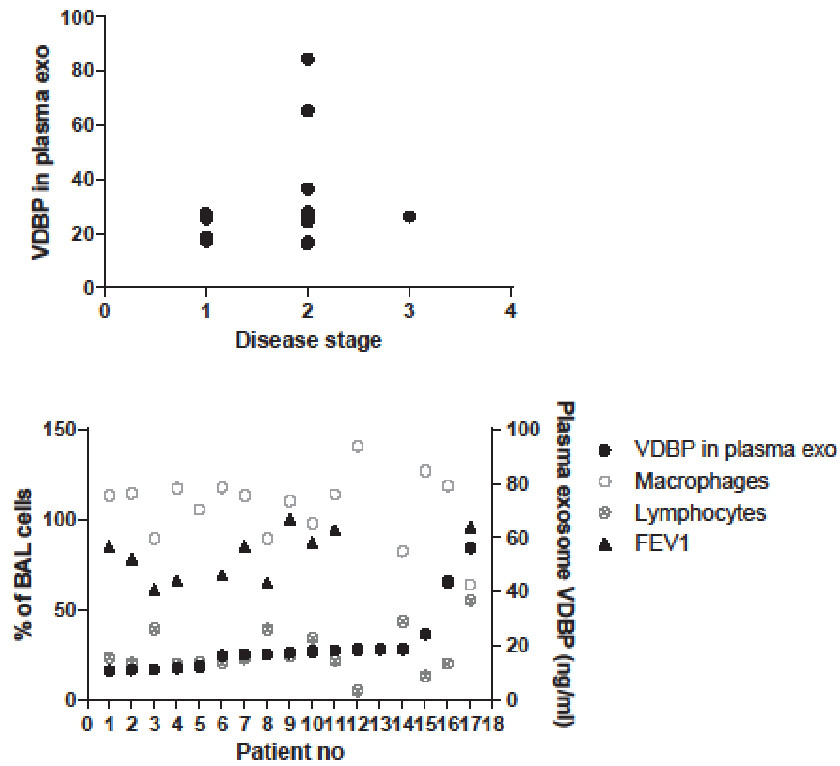
Figure E4: VDBP levels related to disease stages and BAL cell compositions.



The plasma exosome-associated VDBP levels were related to patient disease stage (upper fig), and to the main cellular composition of the BAL fluid as well as the main spirometric readout (FEV1) for each patient (lower fig).

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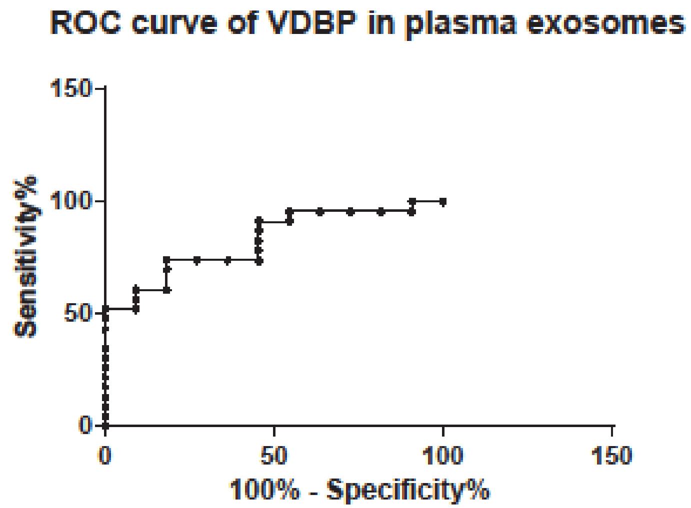
Figure E5: ROC curve of VDBP levels in plasma exosomes.



Based on the levels of VDBP associated to plasma exosomes, and ROC curve was plotted showing an AUC of 0.8261 (p=0.0024) assuming a 95% confidence interval. Assuming a diagnostic test threshold of 24.4 ng/ml exosomal VDBP in serum to discriminate patients from healthy, the sensitivity for the test is 73.9% and the specificity is 81.2%. The positive and negative predictive values are 89% and 60% respectively.

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Online Repository References

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Protein Pilot summary and identified proteins

Excel Work Book Summary

Sheet name	Description
All_values	All protein detected in Sarcoidosis patients and healthy controls
840proteins	Proteins quantified in sarcoidosis samples. The list if aim at including values with sufficient values for statistical evaluation, but allows for up to 33% missing
690_consistent_proteins	Short list of 690 proteins that was filtered to influence the affects of missing values. If the calcualtion included sufficient values, a p-value and error factor was calcualted by ProteinPilot. Without these the values, the numbers can be just based on a few or a single data point, thus no E
Data for significant hits	Data attributes for hits
GO_enrich_UP	GO term enrichment for proteins MORE abundant in the Sarcoidosis BALF exosome samples
GO_enrich_DOWN	GO term enrichment for proteins LESS abundant in the Sarcoidosis BALF exosome samples

ROTS results:
Number of resamplings: 10000
a1: 3.4
a2: 1
Top list size: 80
Reproducibility value: 0.6996537
Z-score: 4.01383

145 rows satisfy the condition.

LABELLING SCHEME

	113	114	115	116	117	118	119	121
Exp1	pool	Exo26ref	Exo24ref	Exo15	Exo6	Exo10	Exo11	Exo3

	113	114	115	116	117	118	119	121
Exp2	pool	Exo23ref	Exo25ref	Exo19	Exo13	Exo4	Exo9	Exo1

	113	114	115	116	117	118	119	121
Exp3	pool	Exo22ref	Exo5	Exo17	Exo7	Exo18	pooled samples	Exo16

Exo 6, Exo 3, Exo 9 and Exo 18 are löfgrens.

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All proteins detected	1962	STATUS: unique peptides, measurements							Protein Score (ProteinPilot)						Sequence Co
All proteins detected >1 peptide	1479														
Protein names	PQ	Accession	count experiments :ID	count experiments :Calcs	measurement quantity	All values	>66% values	>1 peptides	Exp1_Unused	Exp1_Total	Exp2_Unused	Exp2_Total	Exp3_Unused	Exp3_Total	Coverage % max
Serum albumin	P02768	sp|P02768|ALBU	3	3	105	1	1	1	221.72	222.47	194.96	195.66	223.54	224.17	85.710001
Alpha-2-macroglobu	P01023	sp|P01023|A2M	3	3	105	1	1	1	236.23	236.72	226.02	226.6	270.35	271.02	74.6299982
Complement C3 (C3	P01024	sp|P01024|CO3_	3	3	105	1	1	1	295.45	295.45	267.5	267.5	306.11	306.11	75.7099986
Ig kappa chain C regi	P01834	sp|P01834|IGKC	3	3	105	1	1	1	60.34	61.17	61.66	62.53	76.69	77.48	84.9099994
Polymeric immunogl	P01833	sp|P01833|PIGR	3	3	105	1	1	1	121.58	123.71	131.9	133.18	133.66	134.88	65.1799977
Complement C4-B (B	P0C0L5	sp|P0C0L5|CO4B	3	3	105	1	1	1	174.47	177.98	157.65	162.38	194.08	196.23	56.5900028
Ig gamma-1 chain C	P01857	sp|P01857|IGHG	3	3	105	1	1	1	97.27	98.1	79.52	80.1	91.8	92.17	86.6699994
Ig alpha-1 chain C re	P01876	sp|P01876|IGHA	3	3	105	1	1	1	110.65	110.93	104.73	104.94	116.79	117.05	66.8600023
Fibronectin (FN) (Col	P02751	sp|P02751|FINC	3	3	105	1	1	1	175.92	176.06	155.64	156.49	161.39	162.44	49.4100004
Serotransferrin (Tran	P02787	sp|P02787|TRFE	3	3	105	1	1	1	131.38	131.83	111.65	112.24	115.22	116	73.0700016
Ig gamma-4 chain C	P01861	sp|P01861|IGHG	3	3	105	1	1	1	10.42	70.56	6.12	50.62	11.95	63.39	75.8400023
Ig gamma-2 chain C	P01859	sp|P01859|IGHG	3	3	105	1	1	1	36.41	83.06	26.36	66.95	35.01	74.88	84.0499997
Ig alpha-2 chain C re	P01877	sp|P01877|IGHA	3	3	105	1	1	1	23.25	90.27	9.4	78.05	10.55	93.93	71.7599988
Ig gamma-3 chain C	P01860	sp|P01860|IGHG	3	3	105	1	1	1	14.45	78.81	12.44	70.77	10.95	78.89	72.9399979
Galectin-3-binding p	Q08380	sp|Q08380|LG3B	3	3	105	1	1	1	90.96	91.68	85.72	86.42	81.7	82.35	56.9199979
Filamin-A (FLN-A) (A	P21333	sp|P21333|FLNA	3	3	105	1	1	1	170.2	172.68	185.98	187.02	155.81	158.48	46.9999999
Pulmonary surfactan	P35247	sp|P35247|SFTP	3	3	105	1	1	1	60.72	61.31	43.59	43.59	30.04	30.37	66.1300004
Actin, cytoplasmic 2	P63261	sp|P63261|ACTG	3	3	105	1	1	1	53.16	53.54	55.68	55.93	68.43	68.61	61.3300026
IgGFc-binding protei	Q9Y6R7	sp|Q9Y6R7|FCGB	3	3	105	1	1	1	114.57	115.71	118.43	118.71	123.76	124.14	24.7400001
Ig mu chain C region	P01871	sp|P01871|IGHM	3	3	105	1	1	1	70.53	70.99	61.11	62.05	66.84	67.84	62.1699989
Myosin-9 (Cellular m	P35579	sp|P35579|MYH9	3	3	105	1	1	1	121.78	124.64	150.59	152.76	155.16	157.65	45.6099987
Cytoplasmic dynein	Q14204	sp|Q14204|DYHC	3	3	105	1	1	1	162.41	165.09	184.09	185.43	136.1	137.77	22.5600004
Ig heavy chain V-III r	P01766	sp|P01766|HV30	3	3	105	1	1	1	14.86	22.75	24.8	25.25	35.8	36.2	58.3299995
Haptoglobin (Zonulin	P00738	sp|P00738|HPT_	3	3	105	1	1	1	72.73	73.02	59.67	60.07	61.03	61.34	72.9099989
Alpha-1-antitrypsin (	P01009	sp|P01009|A1AT	3	3	105	1	1	1	71.15	73.44	61.86	63.5	68.84	70.55	69.3799973
BPI fold-containing f	Q8TDL5	sp|Q8TDL5|LPLC	3	3	105	1	1	1	65.71	66.56	51.56	52.27	64.85	65.48	77.8900027
Ig kappa chain V-I re	P01608	sp|P01608|KV11	3	3	105	1	1	1	4.01	16.03	3.64	17.66	3.74	18.35	38.8900012
Moesin (Membrane-	P26038	sp|P26038|MOE	3	3	105	1	1	1	49.33	68.72	70.03	70.4	77.24	77.84	63.5999978

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Proteins quantified in sarcoidosis samples. The list if aim at including values with sufficient values for statistical evaluation, but allows for up to 33% missing values*

Count 840 proteins

* these are error factors and p-values from ProteinPilot and not actual ratios

ID	IF	no	countsarc	countkof	countEF	Protein names	PQ	Accession	count_exp.ID	Best	Exp1_Unused	Exp1_Total	Exp2_Unused	Exp2_Total	Exp3_Unused	Exp3_Total	CoverMax	Exp1_Cov95	Exp2_Cov95	Exp3_Cov95	Pmax	Exp1_Peptides	Exp2_Peptides	Exp3_Peptides	AvCalc	CalcMax
A5D8V6	1	2	55	42	80	Vacuolar prote	A5D8V6	sp|A5D8V6|VP	3	4.58	3.13	3.44	4.58	4.69	2.19	2.37	12.1100001	12.1100001	5.63400015	8.73199999	3	3	2	2	2	3
A5YKK6	1	3	55	42	80	CCR4-NOT tran	A5YKK6	sp|A5YKK6|CN	3	4.92	4.52	8.33	4.92	7.38	2.88	4.78	1.30500002	1.30500002	1.30500002	0.42090002	2	2	2	1	2	2
A6NJ16	1	4	55	34	92	Putative V-set	A6NJ16	sp|A6NJ16|VSI	3	7.11	1.73	2.92	2.99	7.71	7.11	8.62	36.590001	33.3299995	24.3900001	36.590001	8	4	6	8	6	13
B0I1T2	1	5	75	50	120	Unconventiona	B0I1T2	sp|B0I1T2|MYI	3	8.21	5.01	8.57	5.38	8.56	8.21	11.65	8.74300003	6.28700033	6.87600002	8.74300003	5	4	5	5	4	6
B9A064	1	6	75	50	120	Immunoglobuli	B9A064	sp|B9A064|IGI	3	35.84	35.84	35.84	27.8	27.8	6.27	31.06	50	50	42.9899991	46.7299998	68	62	68	53	54	80
O00151	1	7	55	42	80	PDZ and LIM d	O00151	sp|O00151|PD	3	5.13	4.49	4.87	5.13	5.3	4	4.13	20.0599998	11.2499997	20.0599998	11.2499997	4	3	4	3	3	5
O00159	1	8	75	50	120	Unconventiona	O00159	sp|O00159|MY	3	56.28	55.39	57.64	56.28	58.93	51.68	56.52	33.3999991	31.6100001	33.3999991	29.0699989	35	31	35	32	35	43
O00160	1	9	75	50	120	Unconventiona	O00160	sp|O00160|MY	3	9.35	7.99	9.98	9.35	10.53	9.11	12.3	5.46399988	5.37299988	5.46399988	5.19100018	6	5	6	5	5	8
O00161	1	10	75	50	120	Synaptosomal-	O00161	sp|O00161|SN	3	6.3	5.14	5.27	6.08	6.27	6.3	6.62	21.7999995	21.7999995	17.5400004	17.5400004	4	4	3	3	7	9
O00182	1	11	75	50	120	Galectin-9 (Gal	O00182	sp|O00182|LEI	3	6	4.05	4.1	6	6.06	3.76	3.82	10.1400003	7.88699985	10.1400003	6.19699992	4	2	4	2	3	4
O00194	1	12	55	34	92	Ras-related pro	O00194	sp|O00194|RB	3	8.68	8.68	10.93	1.87	2.68	2.19	2.28	27.9799998	27.9799998	10.0900002	5.04599996	7	7	4	2	3	4
O00203	1	13	75	50	120	AP-3 complex s	O00203	sp|O00203|AP	3	5.19	5.19	6.78	3.03	3.96	4.44	5.87	3.10800001	2.83400007	0.9141	3.10800001	3	3	1	3	2	3
O00231	1	14	75	50	120	26S proteasom	O00231	sp|O00231|PSI	3	10.47	10.23	11.86	10.47	11.73	9.71	10.74	18.4799999	18.4799999	15.3999999	16.3499996	6	6	6	6	8	11
O00232	1	15	75	50	120	26S proteasom	O00232	sp|O00232|PSI	3	10.49	10.49	11.16	9.57	10.17	8.31	9.08	22.8100002	22.8100002	13.6000007	13.1600007	7	7	5	6	7	8
O00299	1	16	75	50	120	Chloride intrac	O00299	sp|O00299|CLI	3	16.66	13.97	15.01	15.79	17.23	16.66	18.1	56.0199976	56.0199976	44.8100001	48.1299996	12	12	12	11	19	22
O00303	1	17	75	50	120	Eukaryotic tran	O00303	sp|O00303|EIF	3	7.9	7.9	8	6	6.02	6.05	6.2	14.5699993	14.5699993	12.3199999	12.3199999	4	4	4	3	4	5
O00391	1	18	75	50	120	Sulfhydryl oxid	O00391	sp|O00391|QS	3	36.25	28.96	30.8	36.25	37.39	26.45	28.2	30.6600004	25.2999991	30.6600004	24.0999997	24	20	24	17	22	28
O00410	1	19	55	42	80	Importin-5 (Imp	O00410	sp|O00410|IPC	3	6.2	6.2	7.46	6.15	6.85	4.05	5.22	4.28400002	4.28400002	4.01099995	3.19099985	4	4	4	3	4	4
O00560	1	21	75	50	120	Syntenin-1 (Me	O00560	sp|O00560|SD	3	48.12	44.38	45.29	48.12	48.38	46.23	47.96	72.149998	68.4599996	65.7700002	72.149998	70	52	61	70	45	62
O00571	1	22	75	50	120	ATP-dependent	O00571	sp|O00571|DD	3	8.84	2.38	2.89	8.84	9.01	4.77	4.98	6.49499968	5.28700016	6.49499968	5.73999994	4	3	4	3	3	4
O00592	1	23	55	34	92	Podocalyxin (G	O00592	sp|O00592|PO	3	4.01	1.52	1.7	3.89	4	4.01	4.18	3.5840001	1.25399996	3.5840001	3.5840001	2	1	2	2	2	4
O00764	1	26	75	50	120	Pyridoxal kinas	O00764	sp|O00764|PD	3	12.07	12.07	12.21	7.57	7.74	8.73	8.85	25	25	16.99	22.1200004	7	7	4	5	4	6

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Protein Short List

Protein names	PQ	Accession	count experiments ID	count experiments with quantification	measurement quantity	All values	>66% values	>1 peptides	Exp1_Unused	Exp1_Total	Exp2_Unused	Exp2_Total	Exp3_Unused	Exp3_Total	Coverage % max	Exp1_Cov.%
STATUS: unique peptides, calculation consistency, experimental representation									Protein Score						Sequence Coverage	
Serum albumin	P02768	sp|P02768|ALI	3	3	105	1	1	1	221.72	222.47	194.96	195.66	223.54	224.17	85.710001	85.710001
Alpha-2-macro	P01023	sp|P01023|A2I	3	3	105	1	1	1	236.23	236.72	226.02	226.6	270.35	271.02	74.6299982	74.2900014
Complement C	P01024	sp|P01024|CO	3	3	105	1	1	1	295.45	295.45	267.5	267.5	306.11	306.11	75.7099986	75.7099986
Ig kappa chain	P01834	sp|P01834|IG	3	3	105	1	1	1	60.34	61.17	61.66	62.53	76.69	77.48	84.9099994	84.9099994
Polymeric imm	P01833	sp|P01833|PI	3	3	105	1	1	1	121.58	123.71	131.9	133.18	133.66	134.88	65.1799977	57.7199996
Complement C	P0C0L5	sp|P0C0L5|CO	3	3	105	1	1	1	174.47	177.98	157.65	162.38	194.08	196.23	56.5900028	55.8499992
Ig gamma-1 ch	P01857	sp|P01857|IG	3	3	105	1	1	1	97.27	98.1	79.52	80.1	91.8	92.17	86.6699994	86.6699994
Ig alpha-1 chai	P01876	sp|P01876|IG	3	3	105	1	1	1	110.65	110.93	104.73	104.94	116.79	117.05	66.8600023	66.8600023
Fibronectin (FN	P02751	sp|P02751|FIN	3	3	105	1	1	1	175.92	176.06	155.64	156.49	161.39	162.44	49.4100004	49.4100004
Serotransferrin	P02787	sp|P02787|TRI	3	3	105	1	1	1	131.38	131.83	111.65	112.24	115.22	116	73.0700016	73.0700016
Ig gamma-4 ch	P01861	sp|P01861|IG	3	3	105	1	1	1	10.42	70.56	6.12	50.62	11.95	63.39	75.8400023	75.8400023
Ig gamma-2 ch	P01859	sp|P01859|IG	3	3	105	1	1	1	36.41	83.06	26.36	66.95	35.01	74.88	84.0499997	84.0499997
Ig alpha-2 chai	P01877	sp|P01877|IG	3	3	105	1	1	1	23.25	90.27	9.4	78.05	10.55	93.93	71.7599988	71.7599988
Ig gamma-3 ch	P01860	sp|P01860|IG	3	3	105	1	1	1	14.45	78.81	12.44	70.77	10.95	78.89	72.9399979	72.9399979
Galectin-3-binc	Q08380	sp|Q08380|LG	3	3	105	1	1	1	90.96	91.68	85.72	86.42	81.7	82.35	56.9199979	56.9199979
Filamin-A (FLN-	P21333	sp|P21333|FLI	3	3	105	1	1	1	170.2	172.68	185.98	187.02	155.81	158.48	46.9999999	44.6200013
Pulmonary surf	P35247	sp|P35247|SF	3	3	105	1	1	1	60.72	61.31	43.59	43.59	30.04	30.37	66.1300004	66.1300004
Actin, cytoplas	P63261	sp|P63261|AC	3	3	105	1	1	1	53.16	53.54	55.68	55.93	68.43	68.61	61.3300026	56.5299988
IgGFc-binding p	Q9Y6R7	sp|Q9Y6R7|FC	3	3	105	1	1	1	114.57	115.71	118.43	118.71	123.76	124.14	24.7400001	24.3499994
Ig mu chain C r	P01871	sp|P01871|IG	3	3	105	1	1	1	70.53	70.99	61.11	62.05	66.84	67.84	62.1699989	62.1699989
Myosin-9 (Cellu	P35579	sp|P35579|MY	3	3	105	1	1	1	121.78	124.64	150.59	152.76	155.14	157.65	45.6099987	34.3400002
Cytoplasmic dy	Q14204	sp|Q14204|DY	3	3	105	1	1	1	162.41	165.09	184.09	185.43	136.1	137.77	22.5600004	21.6999993
Ig heavy chain	P01766	sp|P01766|HV	3	3	105	1	1	1	14.86	22.75	24.8	25.25	35.8	36.2	58.3299995	51.6700029
Haptoglobin (Z	P00738	sp|P00738|HP	3	3	105	1	1	1	72.73	73.02	59.67	60.07	61.03	61.34	72.9099989	72.9099989
Alpha-1-antitry	P01009	sp|P01009|A1	3	3	105	1	1	1	71.15	73.44	61.86	63.5	68.84	70.55	69.3799973	68.1800008
BPI fold-contai	Q8TDL5	sp|Q8TDL5|LP	3	3	105	1	1	1	65.71	66.56	51.56	52.27	64.85	65.48	77.8900027	68.589999
Ig kappa chain	P01608	sp|P01608|KV	3	3	105	1	1	1	4.01	16.03	3.64	17.66	3.74	18.35	38.8900012	31.4799994
Moesin (Memb	P26038	sp|P26038|MO	3	3	105	1	1	1	49.33	68.72	70.03	70.4	77.24	77.84	63.5999978	52.1700025
Sushi domain-c	Q9UGT4	sp|Q9UGT4|SU	3	3	105	1	1	1	59.76	60.77	58.23	58.63	49.57	50.57	45.0100005	41.3599998
Lactotransferri	P02788	sp|P02788|TRI	3	3	105	1	1	1	67.17	74.66	49.49	53.3	72.77	80.56	63.9400005	63.9400005
Ig kappa chain	P04206	sp|P04206|KV	3	3	105	1	1	1	4	31.33	6	34.3	6	40.13	88.0699992	88.0699992
Keratin, type I	P35527	sp|P35527|K1	3	3	105	1	1	1	59.2	62.45	58.76	60.64	52.69	55.37	57.7799976	55.2200019

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Ras-related C3	P63000	sp|P63000|RA	3	3	105	1	1	1	12.75	15.17	8.74	11.16	2.24	9.05	37.5	37.5
Transmembran	Q72404	sp|Q72404|TN	3	3	105	1	1	1	18.49	18.96	17.07	17.71	16.73	17.33	15.1700005	13.4800002
Tubulin polyme	Q9BW30	sp|Q9BW30|TI	3	3	105	1	1	1	14.67	15.12	16.26	17	18.66	18.73	51.700002	45.449999
Eukaryotic initi	P60842	sp|P60842|IF4	3	3	105	1	1	1	17.05	17.65	17.05	17.16	14.46	14.88	23.3999997	23.3999997
40S ribosomal	P23396	sp|P23396|RS	3	3	105	1	1	1	16.41	17.09	20.73	21.38	14.26	15.04	47.7400005	41.9800013
Phospholipid tr	P55058	sp|P55058|PL	3	3	105	1	1	1	11.21	11.72	12.74	13.3	17.79	18.65	16.8400005	9.53299999
Spectrin alpha	Q13813	sp|Q13813|SP	3	3	105	1	1	1	15.35	20.54	21.31	26.15	20.2	25.32	5.54200001	4.69300002
E3 ubiquitin-pr	Q63HN8	sp|Q63HN8|RI	3	3	105	1	1	1	13.92	20.58	15.78	20.64	12.03	15.15	4.32900004	2.92700008
26S protease r	P35998	sp|P35998|PR	3	3	105	1	1	1	15.81	17.44	13.51	15.44	14.72	16.97	24.0199998	24.0199998
60S ribosomal	P50914	sp|P50914|RL	3	3	105	1	1	1	6.83	7.26	9.75	10.06	6.49	6.72	29.3000013	18.1400001
Eukaryotic tran	P55884	sp|P55884|EIF	3	3	105	1	1	1	14.61	16.55	16.54	17.81	11.8	12.89	12.5300005	11.0600002
Proteasome su	P25787	sp|P25787|PS	3	3	105	1	1	1	12.09	12.35	10.02	12.46	11.13	11.5	36.7500007	35.8999997
Heterogeneous	P61978	sp|P61978|HN	3	3	105	1	1	1	15.95	16.83	18.58	19.29	12.27	13.3	23.33	23.33
Dynactin subur	Q13561	sp|Q13561|DC	3	3	105	1	1	1	4.24	4.29	21.65	22.48	9.82	11.01	29.6799988	7.73100033
T-complex prot	P48643	sp|P48643|TC	3	3	105	1	1	1	12.64	14.65	14.68	18.14	15.6	18.93	19.2200005	12.2000001
26S protease r	P62191	sp|P62191|PR	3	3	105	1	1	1	10.6	14.32	12.4	13.33	12.12	13.21	20.9099993	17.9499999
60S ribosomal	P27635	sp|P27635|RL	3	3	105	1	1	1	9.03	9.26	13.75	14.04	11.49	11.7	38.7899995	22.4299997
Tumor suscept	Q99816	sp|Q99816|TS	3	3	105	1	1	1	9.16	9.31	7.15	7.32	12.75	13.43	24.8699993	14.6200001
Coronin-1C (Co	Q9ULV4	sp|Q9ULV4|CC	3	3	105	1	1	1	16.24	16.69	19.59	19.98	8.54	9.66	18.1400001	16.0300002
Carboxypeptid	P14384	sp|P14384|CB	3	3	105	1	1	1	9.45	10.46	12.23	13.04	10.64	11.47	21.8999997	15.8000007
Vacuolar prote	Q9UN37	sp|Q9UN37|V	3	3	105	1	1	1	14.12	14.73	10.77	16.77	13.69	14.75	19.2200005	19.2200005
Proteasome su	P49721	sp|P49721|PS	3	3	105	1	1	1	11.67	14.36	11.39	12.44	8.12	9.23	41.2900001	41.2900001
Glutamine--fru	Q06210	sp|Q06210|GF	3	3	105	1	1	1	16.9	18.39	7.98	9.05	16.44	19.05	15.5900002	15.5900002
V-type proton	P21281	sp|P21281|VA	3	3	105	1	1	1	6.19	6.38	16.95	17.47	12.74	13.16	28.9600015	7.82800019
Bifunctional pu	P31939	sp|P31939|PU	3	3	105	1	1	1	16.11	16.6	11.8	12.14	12.35	12.81	21.9600007	21.9600007
26S protease r	P62333	sp|P62333|PR	3	3	105	1	1	1	7.72	9.67	15.95	17.73	12.79	14.75	27.5099993	14.6500006
Vacuolar prote	Q709C8	sp|Q709C8|VP	3	3	105	1	1	1	13.15	17.86	17.76	22.99	6.31	8.17	3.94399986	2.34500002
Junction plako	P14923	sp|P14923|PL	3	3	105	1	1	1	11.51	12.67	7.05	7.39	7.48	7.91	13.0199999	13.0199999
G-protein coup	Q9NQ84	sp|Q9NQ84|G	3	3	105	1	1	1	10.88	11.2	6.65	6.71	2.46	2.88	20.86	20.86
Cell division co	P60953	sp|P60953|CD	3	3	105	1	1	1	17.38	17.5	12.23	12.4	10.48	10.68	48.1700003	48.1700003
Protein AMBP	P02760	sp|P02760|AM	3	3	105	1	1	1	11.67	12.11	10.84	11.2	15.74	16.49	28.6900014	18.4699997
Gamma-glutan	P19440	sp|P19440|GG	3	3	105	1	1	1	12.83	13.82	12.59	12.94	16.54	17.04	19.3299994	15.2899995
CD59 glycoprot	P13987	sp|P13987|CD	3	3	105	1	1	1	6.27	6.3	4.38	4.44	5.78	5.88	34.3800008	34.3800008

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5.92	0.12	4.10	0.57	0.49	977.18	0.00	560.79	1.08	0.82	26.69	0.04	28.73	1.13	0.49	4.56	0.25	5.17	0.93	0.79	16.19	0.06	14.99
1.27	0.51	0.83	1.37	0.10	1.48	0.93	2.02	1.62	0.00	1.15	1.41	1.87	1.14	0.06	1.15	0.99	1.32	0.84	0.14	1.28	0.65	1.08
1.16	1.22	1.65	0.96	0.56	1.18	0.81	1.13	1.21	0.01	1.13	1.08	1.37	0.76	0.00	1.15	0.66	0.87	0.98	0.91	1.41	0.70	1.38
1.21	0.88	1.28	1.34	0.16	1.55	0.86	2.08	0.92	0.18	1.14	0.81	1.05	1.10	0.30	1.22	0.90	1.35	1.01	0.86	1.21	0.84	1.22
1.26	0.75	1.20	1.05	0.55	1.22	0.86	1.29	0.75	0.03	1.29	0.58	0.97	1.05	0.68	1.29	0.81	1.35	0.85	0.28	1.38	0.62	1.17
1.14	0.59	0.76	0.83	0.24	1.42	0.58	1.17	1.68	0.00	1.14	1.47	1.91	1.28	0.00	1.14	1.13	1.46	1.06	0.64	1.31	0.81	1.38
1.20	0.98	1.40	1.08	0.39	1.20	0.90	1.30	0.75	0.00	1.17	0.64	0.87	0.87	0.15	1.22	0.71	1.06	0.92	0.16	1.13	0.81	1.04
1.15	0.90	1.18	1.08	0.22	1.15	0.94	1.24	0.74	0.01	1.24	0.60	0.91	0.92	0.18	1.15	0.80	1.05	1.01	0.91	1.27	0.80	1.28
1.25	0.76	1.19	1.21	0.23	1.43	0.85	1.74	0.74	0.03	1.31	0.56	0.96	0.87	0.16	1.23	0.71	1.08	0.94	0.72	1.48	0.64	1.40
1.44	0.69	1.43	0.64	0.10	1.95	0.33	1.26	0.44	0.04	2.07	0.21	0.90	0.50	0.09	2.66	0.19	1.32	0.65	0.04	1.49	0.43	0.96
1.17	0.99	1.35	0.66	0.00	1.18	0.56	0.78	0.67	0.03	1.41	0.47	0.94	0.86	0.17	1.27	0.68	1.09	0.87	0.36	1.43	0.61	1.24
1.30	0.79	1.33	0.71	0.11	1.62	0.44	1.15	0.53	0.02	1.58	0.33	0.83	0.57	0.01	1.41	0.40	0.81	0.66	0.05	1.49	0.44	0.99
1.31	1.14	1.97	1.06	0.64	1.32	0.80	1.40	0.86	0.18	1.28	0.67	1.10	1.04	0.48	1.15	0.91	1.20	0.77	0.08	1.34	0.58	1.04
1.78	0.37	1.17	0.59	0.20	2.49	0.24	1.48	0.58	0.07	1.83	0.32	1.06	0.56	0.07	1.93	0.29	1.07	0.62	0.10	1.81	0.34	1.13
1.12	0.92	1.16	0.89	0.17	1.20	0.74	1.07	0.71	0.01	1.24	0.58	0.88	0.77	0.03	1.26	0.61	0.97	0.72	0.00	1.22	0.59	0.88
1.19	0.92	1.31	0.85	0.29	1.43	0.59	1.21	0.67	0.05	1.47	0.46	0.99	0.75	0.00	1.15	0.65	0.86	0.91	0.17	1.16	0.78	1.06
1.18	1.16	1.61	0.68	0.02	1.34	0.51	0.91	0.59	0.00	1.32	0.45	0.78	0.69	0.01	1.27	0.54	0.87	0.91	0.31	1.25	0.73	1.13
1.20	0.61	0.88	0.71	0.01	1.23	0.58	0.88	1.20	0.01	1.14	1.05	1.36	1.09	0.16	1.14	0.96	1.24	0.85	0.16	1.28	0.66	1.09
1.34	1.13	2.02	0.91	0.34	1.27	0.72	1.16	1.07	0.39	1.20	0.89	1.28	1.19	0.05	1.20	1.00	1.43	0.98	0.79	1.24	0.79	1.21
1.25	0.87	1.35	0.71	0.01	1.26	0.57	0.90	1.23	0.14	1.35	0.91	1.66	1.38	0.00	1.19	1.17	1.64	1.06	0.63	1.36	0.78	1.45
1.41	0.46	0.92	0.55	0.04	1.71	0.32	0.94	1.20	0.05	1.20	1.00	1.45	0.95	0.51	1.20	0.79	1.15	0.75	0.11	1.48	0.51	1.11
1.48	0.42	0.91	0.56	0.07	1.99	0.28	1.11	0.46	0.03	1.89	0.24	0.86	0.46	0.03	1.83	0.25	0.84	0.62	0.05	1.64	0.38	1.02
1.14	1.08	1.42	1.01	0.91	1.20	0.84	1.21	0.88	0.11	1.19	0.74	1.04	0.80	0.10	1.33	0.60	1.06	1.12	0.33	1.30	0.86	1.46
1.38	0.75	1.43	0.80	0.12	1.38	0.58	1.09	0.81	0.41	1.81	0.45	1.47	0.93	0.56	1.37	0.68	1.27	0.81	0.14	1.37	0.59	1.11
1.24	0.85	1.30	0.92	0.45	1.29	0.71	1.19	0.76	0.09	1.39	0.55	1.06	1.06	0.51	1.23	0.86	1.30	0.98	0.86	1.41	0.69	1.38
1.15	0.95	1.27	0.88	0.07	1.15	0.76	1.01	0.80	0.10	1.33	0.60	1.06	0.93	0.45	1.24	0.75	1.15	0.82	0.08	1.26	0.65	1.03
1.69	0.38	1.09	0.76	0.08	1.42	0.54	1.08	0.83	0.45	2.31	0.36	1.92	0.69	0.06	1.49	0.46	1.02	0.76	0.30	2.31	0.33	1.76
1.48	0.85	1.87	0.75	0.15	1.74	0.43	1.30	1.07	0.54	1.48	0.72	1.58	1.06	0.57	1.48	0.72	1.58	1.12	0.81	5.60	0.20	6.27
3.51	0.28	3.44	0.90	0.48	3.51	0.26	3.16	1.05	0.69	3.51	0.30	3.70	0.96	0.74	3.51	0.27	3.36	1.16	0.45	5.07	0.23	5.88
1.30	0.66	1.12	0.98	0.85	1.30	0.76	1.28	1.13	0.24	1.30	0.87	1.47	1.21	0.10	1.30	0.93	1.57	0.96	0.64	1.30	0.74	1.24
1.44	0.71	1.47	1.14	0.16	1.22	0.94	1.40	1.35	0.03	1.29	1.05	1.73	1.58	0.00	1.20	1.31	1.89	0.91	0.64	1.55	0.59	1.42
1.23	0.48	0.72	0.44	0.00	1.30	0.34	0.57	0.44	0.00	1.35	0.32	0.59	2.34	0.00	1.14	2.06	2.67	0.76	0.02	1.24	0.61	0.94
1.54	0.58	1.39	1.01	0.94	1.43	0.70	1.44	1.50	0.13	2.03	0.74	3.04	1.33	0.08	1.43	0.93	1.91	0.67	0.31	3.67	0.18	2.44
1.50	0.70	1.57	0.81	0.03	1.19	0.68	0.97	1.43	0.00	1.17	1.22	1.68	1.03	0.77	1.27	0.81	1.31	0.75	0.05	1.33	0.57	1.00
1.24	0.97	1.50	1.21	0.06	1.22	0.99	1.47	0.87	0.36	1.41	0.62	1.23	0.90	0.41	1.36	0.66	1.22	1.09	0.67	1.67	0.65	1.83
1.36	0.68	1.25	0.89	0.13	1.18	0.75	1.05	1.07	0.33	1.16	0.91	1.24	1.06	0.37	1.17	0.91	1.24	0.97	0.70	1.18	0.83	1.15
1.20	0.73	1.05	0.78	0.23	1.63	0.48	1.28	0.44	0.00	1.41	0.31	0.62	0.50	0.00	1.39	0.36	0.69	0.63	0.01	1.34	0.47	0.84
1.19	0.71	1.00	1.68	0.00	1.20	1.40	2.01	1.21	0.01	1.14	1.06	1.38	1.43	0.00	1.16	1.23	1.66	1.15	0.30	1.34	0.86	1.54
1.38	0.64	1.22	0.92	0.47	1.32	0.70	1.22	1.04	0.63	1.20	0.86	1.24	0.83	0.06	1.22	0.68	1.02	0.99	0.90	1.33	0.74	1.31
1.20	0.78	1.11	0.69	0.00	1.17	0.58	0.80	0.44	0.00	1.31	0.34	0.58	0.46	0.00	1.46	0.32	0.67	0.60	0.01	1.35	0.44	0.80
1.18	0.92	1.28	1.04	0.54	1.14	0.91	1.18	1.15	0.04	1.14	1.01	1.31	1.16	0.03	1.14	1.02	1.32	1.04	0.75	1.29	0.81	1.33
1.17	1.00	1.36	1.14	0.33	1.35	0.84	1.53	1.35	0.00	1.17	1.15	1.58	0.93	0.43	1.24	0.74	1.15	0.83	0.18	1.34	0.62	1.12
1.83	0.53	1.78	0.52	0.25	3.87	0.13	2.00	1.01	0.98	2.81	0.36	2.85	0.69	0.32	2.51	0.27	1.72	0.79	0.56	2.82	0.28	2.22
1.29	0.82	1.35	1.20	0.24	1.45	0.83	1.74	0.64	0.01	1.29	0.49	0.82	0.72	0.08	1.48	0.48	1.06	0.89	0.26	1.28	0.70	1.14
1.31	0.58	1.00	1.02	0.93	1.84	0.56	1.88	1.19	0.07	1.22	0.97	1.46	1.03	0.76	1.25	0.82	1.28	0.92	0.45	1.33	0.69	1.22
1.83	0.40	1.32	0.77	0.32	2.34	0.33	1.81	1.40	0.07	1.52	0.92	2.13	0.86	0.24	1.50	0.57	1.28	0.97	0.76	1.50	0.64	1.45

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STATISTICS — *=multiple hypothesis corrected p-value — RATIOS: (to pooled sample) — Normalization checked log2 ratios (to pooled sample)

ID	ROTS-statistic	*ROTS FDR	log2 Fold Difference	Absolute Fold Difference	Best Protein Score	Coverage %	Unique peptides	Average Calculations	Entry name	Protein names	Gene names	Exo26_ref_114.113_1	Exo24_ref_115.113_1	Exo23_ref_114.113_2	Exo25_ref_115.113_2	Exo22ref_114.113_3
P02748	0.47707635	0	1.59	3.01	22.91	23.9700005	15	21.6666667	CO9_HUMAN	Complement	C9	0.53	0.29	0.32	0.30	0.48
P02760	0.48332254	0	1.56	2.95	15.74	28.6900014	9	19.6666667	AMBP_HUMA	Protein AMBF	AMBP HCP ITI	0.31	0.42	0.41	0.34	0.48
P01042	0.46132829	0	1.53	2.88	19.7	21.1199999	15	13.6666667	KNG1_HUMA	Kininogen-1 (	KNG1 BDK KN	0.40	0.35	0.38	0.40	0.47
P05546	0.44172397	0	1.52	2.86	34.04	42.4800009	24	25.3333333	HEP2_HUMA	Heparin cofac	SERPIND1 HC	0.87	0.29	0.51	0.39	0.41
P09871	0.44162295	0	1.51	2.85	8.7	13.3699998	6	4.66666667	C1S_HUMAN	Complement	C1S	0.26	0.41	0.46	0.38	0.46
P02751	0.44620228	0	1.48	2.79	175.92	49.4100004	191	218.333333	FINC_HUMAN	Fibronectin (F	FN1 FN	0.40	0.39	0.52	0.33	0.61
P18428	0.41915051	0	1.45	2.73	11.18	16.0099998	7	9.66666667	LBP_HUMAN	Lipopolysacch	LBP	0.61	0.32	0.21	0.32	0.70
Q96PD5	0.42072827	0	1.40	2.64	18.55	31.7699999	11	13	PGRP2_HUM	N-acetylmura	PGLYRP2 PGL	0.37	0.45	0.64	0.44	0.45
Q15063	0.47212627	0	1.40	2.64	35.61	32.5399995	28	25	POSTN_HUM	Periostin (PN)	POSTN OSF2	0.25	0.35	0.44	0.26	0.46
P19827	0.4238782	0	1.39	2.62	34.36	24.1500005	23	25.3333333	ITIH1_HUMA	Inter-alpha-tr	ITIH1 IGHEP1	0.47	0.47	0.41	0.40	0.50
P02765	0.40733141	0	1.39	2.61	21.95	43.8699991	16	19	FETUA_HUM	Alpha-2-HS-gl	AHSG FETUA	0.62	0.48	0.43	0.33	0.48
P27169	0.390883	0	1.38	2.60	11.89	25.9200007	6	8.33333333	PON1_HUMA	Serum paraox	PON1 PON	0.35	0.44	0.43	0.66	0.58
P01023	0.39245676	0	1.33	2.52	270.35	74.6299982	416	608.666667	A2MG_HUMA	Alpha-2-macr	A2M CPAMD5	0.26	0.48	0.41	0.45	0.71
P01011	0.39651298	0	1.33	2.51	30.79	47.7499992	30	34.3333333	AACT_HUMA	Alpha-1-antic	SERPINA3 AA	0.68	0.41	0.44	0.35	0.65
P00751	0.40214795	0	1.32	2.50	47.72	38.0899996	34	43.3333333	CFAB_HUMA	Complement	CFB BF BFD	0.45	0.52	0.55	0.36	0.55

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GO term enrichment for proteins MORE abundant in the Sarcoidosis BALF exosome samples

Common GO elements for Proteins more abundant with Löfgrens and non-löfgrens sarcoidosis	Sarc Protein Count	Sarc %	Sarc PValue	Enriched Proteins	Sarc FDR
GOTERM_CC_FAT GO:0005576~extracellular region	77	87.5	2.58E-43	P04196, P01031, P07357, P04114, P07358, Q14624, P05546, P02774, Q9NZP8, P08603, P04217, P36955, P01857, P18428, P01859, P08697, P00738, P01024, P01023, P00734, P55058, P00736, P08637, P02679, P02787, P01042, P02675, P02749, P02748, P07360, P19652, P02743, P02649, P10909, P04275, Q15063, P02790, P00748, P06276, P02647, P00747, P29622, O75636, P02751, P02753, P04003, P19827, P09871, P13671, P19823, P10643, P01011, P05164, P01834, P00751, O43866, P01008, P01009, P00450, P01880, P02652, P02760, P02763, P01860, P02671, P01861, P02768, P02765, P02766, P06727, P43652, P08185, P05156, P27169, Q8N4F0, P04004, Q96PD5, P0C0L5	2.96E-40
GOTERM_CC_FAT GO:0005615~extracellular space	50	56.8182	2.68E-28	P01031, P07357, P07358, P04114, P02774, Q9NZP8, P08603, P36955, P18428, P00738, P08697, P01024, P00734, P01023, P02679, P02787, P01042, P02749, P02675, P19652, P02743, P02649, P10909, P02790, P00748, P00747, P02647, P06276, O75636, P02751, P02753, P13671, P05164, O43866, P01008, P01009, P00450, P02652, P02763, P02671, P02768, P02765, P02766, P06727, P43652, P08185, P27169, P05156, P04004, P0C0L5	3.06E-25
GOTERM_CC_FAT GO:0044421~extracellular region part	52	59.0909	1.01E-26	P01031, P07357, P07358, P04114, P02774, Q9NZP8, P08603, P36955, P18428, P00738, P08697, P01024, P00734, P01023, P02679, P02787, P01042, P02749, P02675, P19652, P02743, P02649, P10909, P04275, Q15063, P02790, P00748, P06276, P02647, P00747, O75636, P02751, P02753, P13671, P05164, O43866, P01008, P01009, P00450, P02652, P02763, P02671, P02768, P02765, P02766, P06727, P43652, P08185, P27169, P05156, P04004, P0C0L5	1.16E-23
GOTERM_BP_FAT GO:0009611~response to wounding	46	52.2727	2.95E-23	O75636, P02751, P01031, P07357, P07358, Q14624, P05546, P04003, Q9NZP8, P08603, P09871, P13671, P01011, P10643, P00751, P18428, P01008, P09960, P01009, P08697, P01024, P02652, P01023, P00734, P00736, P02763, P02679, P02671, P06702, P02787,	4.73E-20
GOTERM_BP_FAT GO:0002526~acute inflammatory response	33	37.5	4.77E-22	O75636, P01031, P02751, P07357, P07358, Q14624, P04003, Q9NZP8, P08603, P09871, P13671, P10643, P01011, P00751, P18428, P01009, P08697, P01024, P00734, P01023, P02652, P00736, P02763, P02787, P02765, P02748, P07360, P19652, P02743, P05156, P10909, P00748, P0C0L5	7.64E-19
GOTERM_BP_FAT GO:0006954~inflammatory response	38	43.1818	1.62E-20	O75636, P02751, P01031, P07357, P07358, Q14624, P04003, Q9NZP8, P08603, P09871, P13671, P10643, P01011, P00751, P18428, P09960, P01009, P08697, P01024, P02652, P01023, P00734, P00736, P02763, P06702, P02787, P01042, P02765, P02748, P07360, P05109, P19652, P02743, P04839, P05156, P10909, P00748, P0C0L5	2.60E-17
GOTERM_BP_FAT GO:0006952~defense response	44	50	4.04E-20	O75636, P02751, P01031, P07357, P07358, Q14624, P04003, Q9NZP8, P08603, P09871, P13671, P10643, P01011, P05164, P00751, O43866, P18428, P09960, P01009, P08697, P00738, P01024, P02652, P01023, P00734, P00736, P02763, P06702, P02787, P01042, P02765, P02748, P07360, P06727, P19652, P05109, P02743, P08311, P04839, P05156, P10909, Q96PD5, P00748, P0C0L5	6.48E-17
GOTERM_BP_FAT GO:0051605~protein maturation by peptide bond	20	22.7273	1.08E-12	O75636, P01031, P07357, P07358, P02749, P04003, P02748, Q9NZP8, P07360, P08603, P09871, P13671, P10643, P00751, P05156, P10909, P01024, P00748, P0C0L5, P00736	1.73E-09
GOTERM_BP_FAT GO:0045087~innate immune response	23	26.1364	7.90E-12	O75636, P01031, P07357, P07358, P04003, P02748, Q9NZP8, P07360, P08603, P06727, P09871, P13671, P10643, P04839, P00751, P18428, P05156, P10909, Q96PD5, P00748, P01024, P0C0L5, P00736	1.26E-08
GOTERM_BP_FAT GO:0050778~positive regulation of immune response	21	23.8636	9.75E-12	O75636, P01031, P30273, P07357, P07358, P04003, P02748, Q9NZP8, P07360, P08603, P09871, P13671, P10643, P00751, P18428, P05156, P10909, P02790, P01024, P0C0L5, P00736	1.56E-08
GOTERM_BP_FAT GO:0006956~complement activation	18	20.4545	1.55E-11	O75636, P01031, P07357, P07358, P04003, P02748, Q9NZP8, P07360, P08603, P09871, P13671, P10643, P00751, P05156, P10909, P01024, P0C0L5, P00736	2.49E-08
GOTERM_BP_FAT GO:0002541~activation of plasma proteins involved in acute inflammatory response	18	20.4545	1.55E-11	O75636, P01031, P07357, P07358, P04003, P02748, Q9NZP8, P07360, P08603, P09871, P13671, P10643, P00751, P05156, P10909, P01024, P0C0L5, P00736	2.49E-08
GOTERM_BP_FAT GO:0016485~protein processing	20	22.7273	2.39E-11	O75636, P01031, P07357, P07358, P02749, P04003, P02748, Q9NZP8, P07360, P08603, P09871, P13671, P10643, P00751, P05156, P10909, P01024, P00748, P0C0L5, P00736	3.82E-08
GOTERM_BP_FAT GO:0051604~protein maturation	20	22.7273	5.85E-11	O75636, P01031, P07357, P07358, P02749, P04003, P02748, Q9NZP8, P07360, P08603, P09871, P13671, P10643, P00751, P05156, P10909, P01024, P00748, P0C0L5, P00736	9.36E-08
GOTERM_MF_FAT GO:0004866~endopeptidase inhibitor activity	19	21.5909	8.99E-11	P04196, P01031, P01042, Q14624, P05546, P02765, P19827, P19823, P01011, P36955, P08185, P01008, P01009, P08697, P01024, P01023, P02760, P29622, P0C0L5	1.13E-07
GOTERM_MF_FAT GO:0030414~peptidase inhibitor activity	19	21.5909	2.14E-10	P04196, P01031, P01042, Q14624, P05546, P02765, P19827, P19823, P01011, P36955, P08185, P01008, P01009, P08697, P01024, P01023, P02760, P29622, P0C0L5	2.69E-07
GOTERM_BP_FAT GO:0048584~positive regulation of response to stimulus	21	23.8636	2.44E-10	O75636, P01031, P30273, P07357, P07358, P04003, P02748, Q9NZP8, P07360, P08603, P09871, P13671, P10643, P00751, P18428, P05156, P10909, P02790, P01024, P0C0L5, P00736	3.91E-07
GOTERM_BP_FAT GO:0006955~immune response	32	36.3636	3.98E-10	O75636, P01031, P07357, P30273, P07358, P04003, Q9NZP8, P08603, P09871, P13671, P10643, P01834, P00751, P01857, P18428, P01859, P01024, P01880, P00736, P08637, P01860, P01861, P02748, P07360, P06727, P08311, P04839, P05156, P10909, P04004, Q96PD5, P00748, P0C0L5	6.38E-07
GOTERM_BP_FAT GO:0006959~humoral immune response	18	20.4545	8.36E-10	O75636, P01031, P07357, P07358, P04003, P02748, Q9NZP8, P07360, P08603, P09871, P13671, P10643, P00751, P05156, P10909, P01024, P0C0L5, P00736	1.34E-06
GOTERM_BP_FAT GO:0002253~activation of immune response	18	20.4545	8.36E-10	O75636, P01031, P07357, P07358, P04003, P02748, Q9NZP8, P07360, P08603, P09871, P13671, P10643, P00751, P05156, P10909, P01024, P0C0L5, P00736	1.34E-06
GOTERM_BP_FAT GO:0002684~positive regulation of immune system process	21	23.8636	9.64E-10	O75636, P01031, P30273, P07357, P07358, P04003, P02748, Q9NZP8, P07360, P08603, P09871, P13671, P10643, P00751, P18428, P05156, P10909, P02790, P01024, P0C0L5, P00736	1.54E-06
GOTERM_BP_FAT GO:0006958~complement activation, classical pathway	15	17.0455	4.16E-09	P01031, P07357, P07358, P04003, P02748, Q9NZP8, P07360, P09871, P13671, P10643, P05156, P10909, P01024, P0C0L5, P00736	6.66E-06
GOTERM_BP_FAT GO:0002455~humoral immune response mediated by circulating immunoglobulin	15	17.0455	4.16E-09	P01031, P07357, P07358, P04003, P02748, Q9NZP8, P07360, P09871, P13671, P10643, P05156, P10909, P01024, P0C0L5, P00736	6.66E-06
GOTERM_BP_FAT GO:0016064~immunoglobulin mediated immune response	16	18.1818	4.80E-09	P01031, P30273, P07357, P07358, P04003, P02748, Q9NZP8, P07360, P09871, P13671, P10643, P05156, P10909, P01024, P0C0L5, P00736	7.69E-06
GOTERM_BP_FAT GO:0019724~B cell mediated	16	18.1818	4.80E-09	P01031, P30273, P07357, P07358, P04003, P02748, Q9NZP8, P07360, P09871, P13671, P10643, P05156, P10909, P01024, P0C0L5, P00736	7.69E-06

The protein count represents the number of proteins matching the term.
The % is the percentatge of the list submitted for the query that was used.
The FDRs are calculatd as percentages

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GO term enrichment for proteins LESS abundant in the Sarcoidosis BALF exosome samples

Common GO elements for Proteins less abundant with Löfgrens and non-löfgrens sarcoidosis	Sarc Protein Count	Sarc %	Sarc PValue	Sarc Proteins	Sarc FDR
GOTERM_MF_FAT GO:0004298~threonine-type endopeptidase activity	12	24.0	1.5E-11	P20618, P28074, P25788, P25789, P28070, P25786, P25787, P40306, P60900, P49721, P49720, P28066	0.00
GOTERM_MF_FAT GO:0070003~threonine-type peptidase activity	12	24.0	1.5E-11	P20618, P28074, P25788, P25789, P28070, P25786, P25787, P40306, P60900, P49721, P49720, P28066	0.00
GOTERM_CC_FAT GO:0005839~proteasome core complex	12	24.0	3.7E-11	P20618, P28074, P25788, P25789, P28070, P25786, P25787, P40306, P60900, P49721, P49720, P28066	0.00
GOTERM_BP_FAT GO:0007049~cell cycle	20	40.0	6.3E-10	Q99816, P25788, P25789, P25786, O75351, P25787, P40306, Q13561, Q8WUM4, Q9UN37, P20618, P28074, P28070, Q14203, Q7LBR1, P60900, P49721, P49720, P53990, P28066	0.00
GOTERM_BP_FAT GO:0031400~negative regulation of protein	14	28.0	1.1E-06	P27105, P25788, P25789, P25786, P25787, P40306, Q03135, P28074, P20618, P28070, P60900, P49721, P49720, P28066	0.00
GOTERM_BP_FAT GO:0032269~negative regulation of cellular protein	15	30.0	1.4E-06	P60228, P27105, P25788, P25789, P25786, P25787, P40306, Q03135, P28074, P20618, P28070, P60900, P49721, P49720, P28066	0.00
GOTERM_BP_FAT GO:0022402~cell cycle process	15	30.0	1.4E-06	Q99816, P25788, P25789, P25786, P25787, P40306, Q13561, P20618, P28074, P28070, Q14203, P60900, P49721, P49720, P28066	0.00
GOTERM_BP_FAT GO:0051248~negative regulation of protein metabolic	15	30.0	1.9E-06	P60228, P27105, P25788, P25789, P25786, P25787, P40306, Q03135, P28074, P20618, P28070, P60900, P49721, P49720, P28066	0.00
GOTERM_BP_FAT GO:0006511~ubiquitin-dependent protein catabolic	13	26.0	3.2E-06	Q99816, P25788, P25789, P25786, P25787, P40306, P28074, P20618, P28070, P60900, P49721, P49720, P28066	0.00
GOTERM_BP_FAT GO:0000278~mitotic cell cycle	14	28.0	4E-06	P25788, P25789, P25786, P25787, P40306, Q13561, P28074, P20618, P28070, Q14203, P60900, P49721, P49720, P28066	0.01
GOTERM_BP_FAT GO:0010605~negative regulation of macromolecule	16	32.0	4.3E-06	P60228, Q99816, P27105, P25788, P25789, P25786, P25787, P40306, Q03135, P20618, P28074, P28070, P60900, P49721, P49720, P28066	0.01
GOTERM_BP_FAT GO:0051444~negative regulation of ubiquitin-protein	12	24.0	4.5E-06	P20618, P28074, P25788, P25789, P28070, P25786, P25787, P40306, P60900, P49721, P49720, P28066	0.01
GOTERM_BP_FAT GO:0051437~positive regulation of ubiquitin-protein	12	24.0	4.5E-06	P20618, P28074, P25788, P25789, P28070, P25786, P25787, P40306, P60900, P49721, P49720, P28066	0.01
GOTERM_BP_FAT GO:0051436~negative regulation of ubiquitin-protein	12	24.0	4.5E-06	P20618, P28074, P25788, P25789, P28070, P25786, P25787, P40306, P60900, P49721, P49720, P28066	0.01
GOTERM_BP_FAT GO:0031145~anaphase-promoting complex-dependent	12	24.0	4.5E-06	P20618, P28074, P25788, P25789, P28070, P25786, P25787, P40306, P60900, P49721, P49720, P28066	0.01
GOTERM_BP_FAT GO:0051352~negative regulation of ligase activity	12	24.0	4.5E-06	P20618, P28074, P25788, P25789, P28070, P25786, P25787, P40306, P60900, P49721, P49720, P28066	0.01
GOTERM_BP_FAT GO:0051439~regulation of ubiquitin-protein ligase activity	12	24.0	4.5E-06	P20618, P28074, P25788, P25789, P28070, P25786, P25787, P40306, P60900, P49721, P49720, P28066	0.01

The protein count represents the number of proteins matching the term.
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The FDRs are calculatd as percentages

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GOTERM_BP_FAT GO:0031397~negative regulation of protein	12	24.0	4.5E-06	P20618, P28074, P25788, P25789, P28070, P25786, P25787, P40306, P60900, P49721, P49720, P28066	0.01
GOTERM_BP_FAT GO:0043632~modification-dependent macromolecule	14	28.0	5.4E-06	Q99816, P25788, P25789, P25786, P25787, P40306, Q8IX04, P20618, P28074, P28070, P60900, P49721, P49720, P28066	0.01
GOTERM_BP_FAT GO:0019941~modification-dependent protein catabolic	14	28.0	5.4E-06	Q99816, P25788, P25789, P25786, P25787, P40306, Q8IX04, P20618, P28074, P28070, P60900, P49721, P49720, P28066	0.01
GOTERM_BP_FAT GO:0051340~regulation of ligase activity	12	24.0	6.5E-06	P20618, P28074, P25788, P25789, P28070, P25786, P25787, P40306, P60900, P49721, P49720, P28066	0.01
GOTERM_BP_FAT GO:0031396~regulation of protein ubiquitination	12	24.0	6.5E-06	P20618, P28074, P25788, P25789, P28070, P25786, P25787, P40306, P60900, P49721, P49720, P28066	0.01
GOTERM_BP_FAT GO:0051443~positive regulation of ubiquitin-protein	12	24.0	6.5E-06	P20618, P28074, P25788, P25789, P28070, P25786, P25787, P40306, P60900, P49721, P49720, P28066	0.01
GOTERM_BP_FAT GO:0051351~positive regulation of ligase activity	12	24.0	6.5E-06	P20618, P28074, P25788, P25789, P28070, P25786, P25787, P40306, P60900, P49721, P49720, P28066	0.01
GOTERM_BP_FAT GO:0031398~positive regulation of protein	12	24.0	6.5E-06	P20618, P28074, P25788, P25789, P28070, P25786, P25787, P40306, P60900, P49721, P49720, P28066	0.01
GOTERM_BP_FAT GO:0051438~regulation of ubiquitin-protein ligase activity	12	24.0	6.5E-06	P20618, P28074, P25788, P25789, P28070, P25786, P25787, P40306, P60900, P49721, P49720, P28066	0.01
GOTERM_CC_FAT GO:0000502~proteasome complex	12	24.0	6.8E-06	P20618, P28074, P25788, P25789, P28070, P25786, P25787, P40306, P60900, P49721, P49720, P28066	0.01
GOTERM_BP_FAT GO:0031401~positive regulation of protein	13	26.0	8.5E-06	P25788, P25789, P25786, P25787, P40306, Q03135, P28074, P20618, P28070, P60900, P49721, P49720, P28066	0.01
GOTERM_BP_FAT GO:0044265~cellular macromolecule catabolic	15	30.0	1.2E-05	P60228, Q99816, P25788, P25789, P25786, P25787, P40306, Q8IX04, P20618, P28074, P28070, P60900, P49721, P49720, P28066	0.02
GOTERM_BP_FAT GO:0044257~cellular protein catabolic process	14	28.0	1.6E-05	Q99816, P25788, P25789, P25786, P25787, P40306, Q8IX04, P20618, P28074, P28070, P60900, P49721, P49720, P28066	0.02
GOTERM_BP_FAT GO:0051603~proteolysis involved in cellular protein	14	28.0	1.6E-05	Q99816, P25788, P25789, P25786, P25787, P40306, Q8IX04, P20618, P28074, P28070, P60900, P49721, P49720, P28066	0.02
GOTERM_BP_FAT GO:0031399~regulation of protein modification process	14	28.0	1.6E-05	P27105, P25788, P25789, P25786, P25787, P40306, Q03135, P28074, P20618, P28070, P60900, P49721, P49720, P28066	0.02
GOTERM_BP_FAT GO:0043161~proteasomal ubiquitin-dependent protein	12	24.0	1.8E-05	P20618, P28074, P25788, P25789, P28070, P25786, P25787, P40306, P60900, P49721, P49720, P28066	0.03
GOTERM_BP_FAT GO:0010498~proteasomal protein catabolic process	12	24.0	1.8E-05	P20618, P28074, P25788, P25789, P28070, P25786, P25787, P40306, P60900, P49721, P49720, P28066	0.03
GOTERM_MF_FAT GO:0004175~endopeptidase activity	12	24.0	2E-05	P20618, P28074, P25788, P25789, P28070, P25786, P25787, P40306, P60900, P49721, P49720, P28066	0.02
GOTERM_BP_FAT GO:0030163~protein catabolic process	14	28.0	2.1E-05	Q99816, P25788, P25789, P25786, P25787, P40306, Q8IX04, P20618, P28074, P28070, P60900, P49721, P49720, P28066	0.03

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GOTERM_BP_FAT GO:0009057~macromolecule catabolic process	15	30.0	2.4E-05	P60228, Q99816, P25788, P25789, P25786, P25787, P40306, Q8IX04, P20618, P28074, P28070, P60900, P49721, P49720, P28066	0.04
GOTERM_BP_FAT GO:0051247~positive regulation of protein metabolic	13	26.0	2.7E-05	P25788, P25789, P25786, P25787, P40306, Q03135, P28074, P20618, P28070, P60900, P49721, P49720, P28066	0.04
GOTERM_BP_FAT GO:0032270~positive regulation of cellular protein	13	26.0	2.7E-05	P25788, P25789, P25786, P25787, P40306, Q03135, P28074, P20618, P28070, P60900, P49721, P49720, P28066	0.04
GOTERM_MF_FAT GO:0070011~peptidase activity, acting on L-amino acid peptides	13	26.0	3E-05	P25788, P25789, P25786, P25787, P40306, P28074, P20618, P28070, P60900, P28838, P49721, P49720, P28066	0.03
GOTERM_BP_FAT GO:0010604~positive regulation of macromolecule	14	28.0	3.4E-05	Q9UM54, P25788, P25789, P25786, P25787, P40306, Q03135, P28074, P20618, P28070, P60900, P49721, P49720, P28066	0.05
GOTERM_BP_FAT GO:0043086~negative regulation of catalytic activity	13	26.0	5.7E-05	P25788, P25789, P25786, P25787, P40306, Q03135, P28074, P20618, P28070, P60900, P49721, P49720, P28066	0.08
GOTERM_MF_FAT GO:0008233~peptidase activity	13	26.0	6E-05	P25788, P25789, P25786, P25787, P40306, P28074, P20618, P28070, P60900, P28838, P49721, P49720, P28066	0.07
GOTERM_BP_FAT GO:0032268~regulation of cellular protein metabolic	15	30.0	0.0001	P60228, P27105, P25788, P25789, P25786, P25787, P40306, Q03135, P28074, P20618, P28070, P60900, P49721, P49720, P28066	0.15
GOTERM_BP_FAT GO:0044092~negative regulation of molecular	13	26.0	0.00014	P25788, P25789, P25786, P25787, P40306, Q03135, P28074, P20618, P28070, P60900, P49721, P49720, P28066	0.20
GOTERM_CC_FAT GO:0019773~proteasome core complex, alpha-subunit	4	8.0	0.00149	P25789, P25787, P60900, P28066	1.78
GOTERM_BP_FAT GO:0043085~positive regulation of catalytic activity	13	26.0	0.00168	P25788, P25789, P25786, P25787, P40306, Q03135, P28074, P20618, P28070, P60900, P49721, P49720, P28066	2.43
GOTERM_BP_FAT GO:0006508~proteolysis	16	32.0	0.0026	P25786, P25787, P40306, Q8IX04, P20618, P28074, P28070, P08174, P60900, P28838,	3.68

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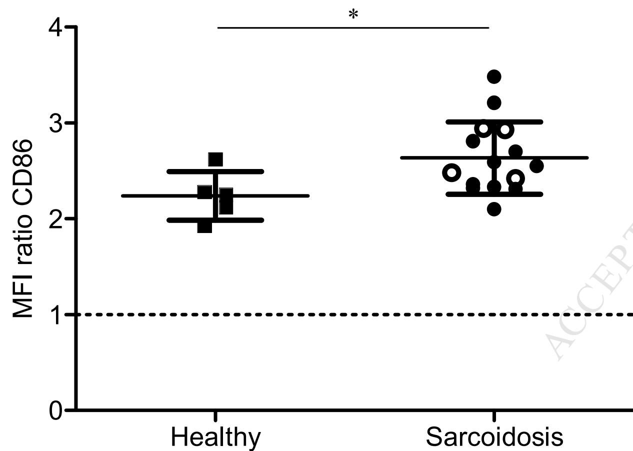

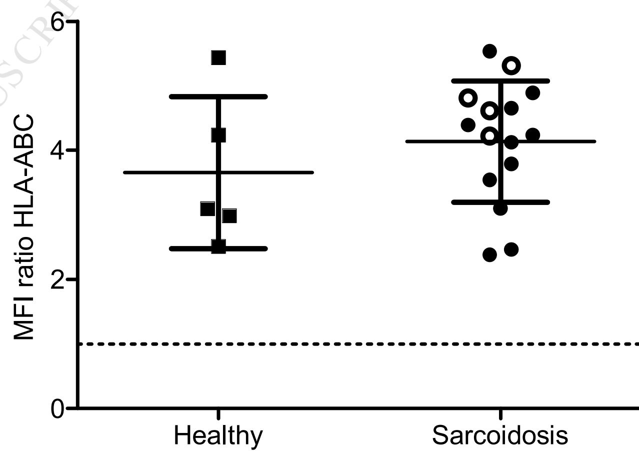

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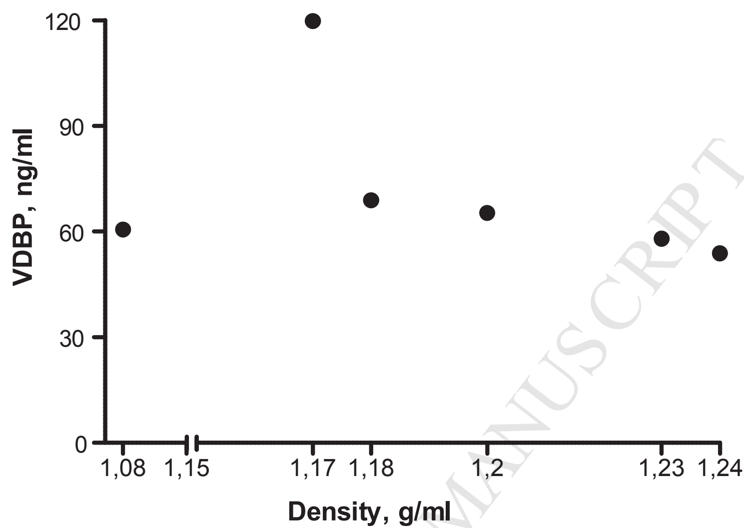

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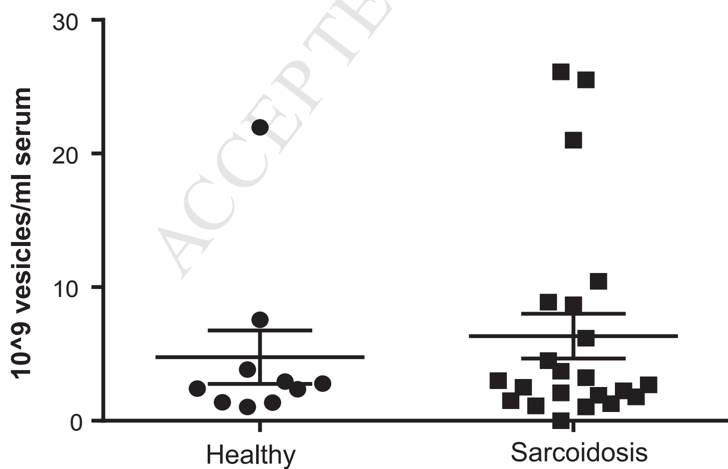

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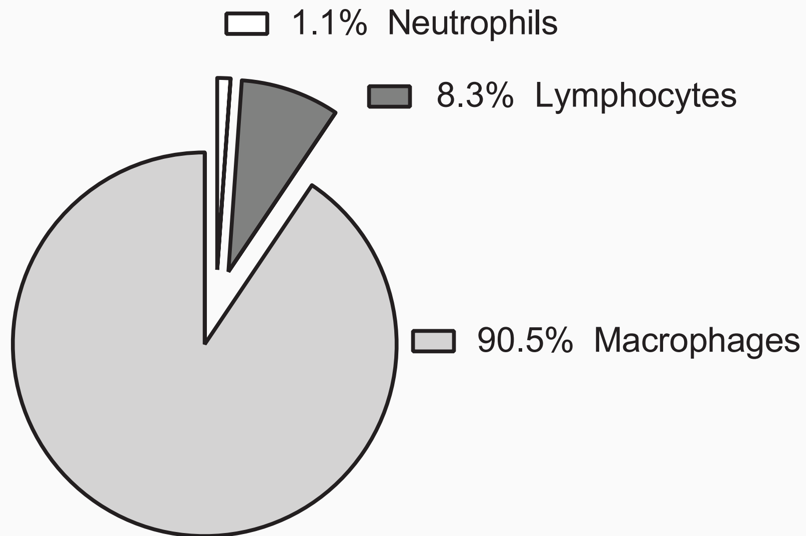

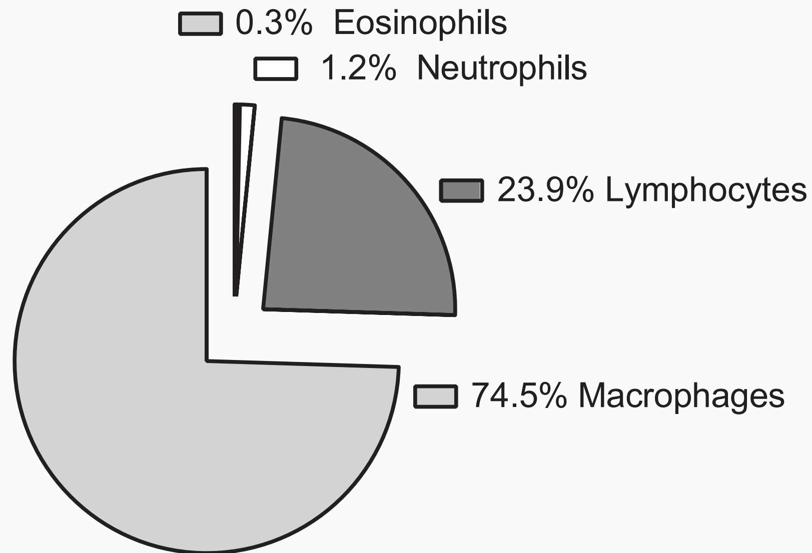

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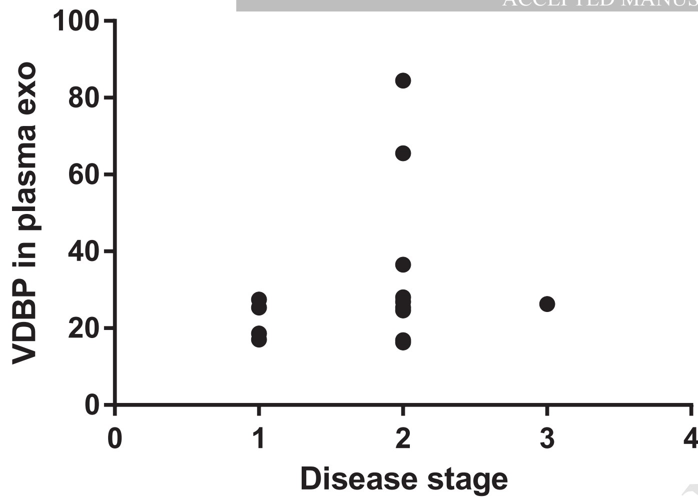

150
100
50
0
% of BAL cells
100
80
60
40
20
0
Plasma exosome VDBP (ng/ml)
0 1 2 3 4 5 6 7 8 9 10 11 12 13 14 15 16 17 18
Patient no
VDBP in plasma exo
Macrophages
Lymphocytes
FEV1

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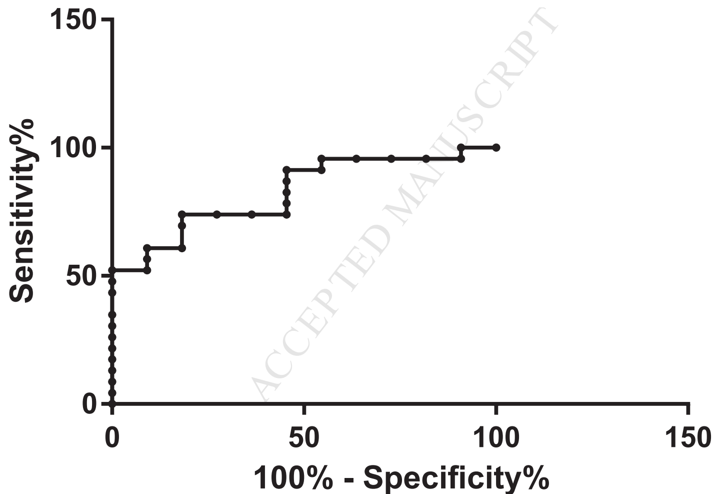

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