Mannose binding lectin in patients with pulmonary tuberculosis: Active and inactive

Mahmoud M. Alsalahya, Gehan F. Almehya, Rasha M. Hendya,⇑ Rasha S. Mohammadb, Yasser Mahmoud Mohammadc

aChest Department, Benha University Hospitals, Egypt
bClinical Pathology Department, Benha University Hospitals, Egypt
cTanta Chest Hospital, Egypt

ABSTRACT

This study was done to assess the relation between serum mannose binding lectin (MBL) and disease activity and extension in patients with tuberculosis.

Methods: The study included 50 pulmonary TB cases recruited from chest department at El Abasia and Tanta Chest Hospitals and 27 patients with other respiratory infections as a positive control group. Also 13 healthy subjects were included as a negative control group. Patients were classified as follows: 30 with active TB (group I), 20 with inactive TB (group II) and 27 with non-TB respiratory diseases as positive control group (group III). Full clinical evaluation, plain chest X-ray (postero-anterior & lateral views), routine laboratory investigations were done to all patients. MBL measurement in serum of all subjects was done by an ELISA assay.

Results: No significant difference in MBL levels seen between males and females in all studied groups. MBL levels were significantly higher in group I & II than the other groups. MBL levels were non-significantly higher in active TB (group I) than inactive TB (group II) while they were significantly higher in active TB cases than in infectious respiratory diseases other than TB and non-infectious respiratory diseases. Levels were non-significantly higher in active TB cases than in controls. MBL levels were significantly higher in TB cases than in infectious respiratory diseases other than TB. 60% of active TB group has high MBL level versus 13.3% of non-TB respiratory infection cases and 8.3% of noninfectious respiratory diseases group. Only one patient with far advanced tuberculosis showed significantly higher levels of MBL than patients with minimal and moderate disease while no significant difference seen between patients with minimal and moderately advanced diseases.

Conclusion: MBL is possibly related to increased susceptibility to tuberculosis. Its levels were nearly the same in both active and inactive tuberculous patients. There is a direct relation between levels of MBL and disease extension.

© 2017 The Egyptian Society of Chest Diseases and Tuberculosis. Production and hosting by Elsevier B.V.

This is an open access article under the CC BY-NC-ND license.

http://dx.doi.org/10.1016/j.ejcdt.2016.12.012

0422-7638/© 2017 The Egyptian Society of Chest Diseases and Tuberculosis. Production and hosting by Elsevier B.V.

This is an open access article under the CC BY-NC-ND license.
asymptomatic, latent infection, and about one in ten latent infections eventually progress to active disease, which, if left untreated, kills more than 50% of those infected cases [3].

Aim of the work

To study the relation between serum MBL and disease activity and extension in patient with tuberculosis.

Subjects and methods

The study included 50 pulmonary TB cases recruited from chest department at El Abasia and Tanta chest hospital and 27 patients with other respiratory infections as a positive control group. Also 13 healthy subjects were included as a negative control group.

Patients were classified into 3 groups: Group I (30 with active TB), group II (20 with inactive TB), group III (27 with non-TB respiratory diseases as positive control group) which was divided into two subgroups: 15 with respiratory infection other than TB (group IIIa) and 12 with non-infectious respiratory diseases (group IIIb). Group IV included 13 apparently healthy subjects as a negative control group.

Exclusion criteria: Patients with liver failure, renal failure, cardiac failure malnutrition diseases, collagen diseases and other systemic diseases like diabetes mellitus were excluded due to possible effect on MBL production.

Included patients were subjected to: Full clinical evaluation (history and examination), plain chest X-ray (postero-anterior & lateral views) plus routine laboratory investigations (CBC, ESR, and liver and renal function tests).

Disease activity was assessed by clinical symptoms, high ESR, soft tissue opacity or cavitation on X-ray and a positive Ziehl Nelson stain smear [3]. Active TB patients were classified according to extent in X-ray into: Minimal, moderately and far advanced [4].

MBL levels were assessed in serum of all subjects by an ELISA assay according to the manufacturer instructions using Human MBL ELISA, Biotechnology Company, USA, CA 94089 kits.

Statistical analysis [5]

Statistical presentation and analysis of the collected data were conducted, using the mean, standard deviation, analysis of variance [ANOVA] test and chi-square test by the SPSS statistical software version 11 for windows. Differences were considered significant when p value is <0.05.

Results

Lowest mean of age was in active TB patients (group I), while the highest was in non-infectious respiratory diseases group (group IIIb). The majority of cases in groups I, II, IIIb were males (90%, 80% & 53.3 respectively) while in groups IIIa, and IV the majority were females (91.7 & 69.2 respectively). Over all males were 62.22% of studied subjects while females were 37.78%.

MBL levels in the studied groups had no significant difference between males and females in all groups and its levels were significantly higher in group I & II than the other groups. Control group showed significantly higher MBL levels than non-TB respiratory infection group and non-infectious respiratory diseases groups. MBL levels were non-significantly higher in active TB (group I) than inactive TB (group II). MBL levels were also significantly higher in active TB cases (group I) than in infectious respiratory diseases other than TB (group IIIb) and in active TB cases (group I) than in non-infectious respiratory diseases (group IIIb). Levels were non-significantly higher in active TB cases (group I) than in controls (group IV). TB cases (group I and II) had significantly higher levels than in infectious respiratory diseases other than TB (group IIIa). TB Cases (group I and II) had non-significantly higher levels of MBL than in controls (group IV).

60% of active TB group had higher MBL levels versus 13.3% of non-TB respiratory infection cases. Normal to low levels were seen in 3.3% of active TB cases and in 40% of non-TB infections cases. MBL was deficient in 36.7% in active TB group versus 46.60% of non-TB infection cases. Differences within groups were statistically significant. 60% of active TB group had high MBL levels versus 23.1% of control group. Normal to low levels were seen in 3.3% of active TB cases and in 69.2% of controls. It was deficient in 36.7% in active TB group compared with 7.7% in the control group. There was statistically significant difference in MBL levels between a patient with extensive disease and patients with both minimal and moderately advanced diseases while no significant difference seen between patients with minimal and moderate diseases.

There was a significant direct correlation between age in years and MBL in μg/ml in group I (active TB) only, while in other groups the relation was non-significant. There was no significant correlation between temperature and MBL in group I (active TB). There was no significant correlation between ESR and MBL in all respiratory infection groups. The correlation was negative in group I while in other groups it was positive.

Discussion

This study aimed at evaluation of MBL level in blood of active (30) & inactive (20) TB cases to evaluate its role in susceptibility to TB as well as the relation between its level and disease extension in patients attending El Abasia and Tanta Chest Hospitals.

Although our TB patients were selected randomly, yet the average age of patients with active & inactive pulmonary TB was around 35–45 years (Table 1 and Fig. 1) meaning that TB infection is more common in the productive age group which is a known fact in TB where Global Tuberculosis Report (2014) [6] stated that about 70% of TB cases occur among the age group of 15–54 years (most productive age group), whereas in the developed countries, elderly people are more susceptible.

In our study, no significant correlation was seen between age and MBL in all groups, except in active TB cases (group I) where it was significant and direct (Table 6). Many studies assessed age dependent MBL alterations and its relation to increase susceptibility to infections [7] and found no relation between MBL levels and age but found a direct relation with alteration in its molecule (genomic mutation) which was associated with increased susceptibility to invasive meningococcal infection. Similarly, Singh et al. (2008) [8] showed that mutations in MBL were associated with rapid progression of disease in HIV infected children and were directly age dependent. In Vietnam in 2014, Hijjkat et al. [9] found an age dependent association of MBL gene polymorphism and the development of pulmonary tuberculosis. It is possible in this study that age dependent increase in MBL levels in active TB patients might be related to gene mutations which resulted in elevated levels of useless molecules [10] (see Table 7).

In the current work, number of males was higher in both active & inactive Pulmonary TB than females (Table 1 and Fig. 2). This result is supported by WHO (2014) report which stated that males had a higher prevalence than females [6]. This higher number of TB in males appears to be the result of a true difference in disease occurrence rather than a difference in access to TB diagnosis and treatment, and could be attributed to either behavioral activity like tobacco smoking (goza and cigarettes). Also, socioeconomic differences play a role as males are at higher risk of exposure to infection due to work and stress. It is possible that all these factors interact.

In the present work, MBL levels showed no significant difference between males and females in all groups as well as totally (Table 2 and Fig. 3), meaning that MBL levels are not affected by gender. This result agreed with Huang et al. (2015) [11] who found no relation between sex and MBL levels in patients with respiratory infections (see Fig. 4).

In this study, it was found that MBL levels were significantly higher in active and inactive TB than other groups (Table 3), with about 60% of tuberculous cases had high level of MBL (Table 5). This means that elevations in MBL could be an immune response to halt access of invading bacilli, or just an acute phase reaction phenomenon. Many studies have assessed TB dependent MBL alterations and its relation to increase of infectivity. In a study by Danish Central Ethical Committee, Selvaraj et al. (2006) [12] found that high MBL levels in blood contributed to increase of susceptibility to tuberculous infections. However, other studies found that MBL is elevated in active tuberculous infection as a part of an acute-phase reaction [13]. Selvaraj et al. (2006) [12] found that despite decrease in M. tuberculosis phagocytosis in wild-type MBL patients with the disease there was higher MBL levels than healthy controls and that patients with wild-type MBL2 genotypes have been generally found to have raised MBL levels in acute phase,
reflecting malfunctioning molecules. Denholm et al. (2010) [13] concluded that MBL is not involved in tuberculosis infection and elevated MBL levels seen in patients with TB represent an acute-phase response. In support of acute phase response is the 1.5–3 times elevations in MBL levels found by Thiel et al. (1991) [14] in post-operative patients (see Table 4).

In the current work, about 36% of active TB cases had deficiency in MBL level (Table 5), this means that deficient MBL could be a risk factor for developing active TB disease, so, both elevated malfunctioning MBL and its deficiency might be responsible for active TB disease. This result is in agreement with Denholm et al. (2010) [13], who reported that patients with high-expression genotypes and genotypes conferring MBL deficiency are not protected against TB. Also, Chen et al. (2015) [15] showed that Promoter -221(Y/X) mutation (YX, XX) could lead to decreased serum MBL and enhanced susceptibility to TB.

Table 3
Comparison of MBL levels among different groups included in the study.

<table>
<thead>
<tr>
<th>Group</th>
<th>MBL (M ± SD)</th>
<th>t test</th>
<th>p</th>
</tr>
</thead>
<tbody>
<tr>
<td>Active vs. Inactive TB groups</td>
<td>Active TB</td>
<td>1.85 ± 1.44</td>
<td>0.55</td>
</tr>
<tr>
<td></td>
<td>Inactive TB</td>
<td>1.64 ± 1.00</td>
<td></td>
</tr>
<tr>
<td>Active TB vs. non-TB infectious group</td>
<td>Active TB</td>
<td>1.85 ± 1.44</td>
<td>2.95</td>
</tr>
<tr>
<td></td>
<td>Non-TB infectious group</td>
<td>0.57 ± 0.51</td>
<td></td>
</tr>
<tr>
<td>Active TB vs. non-infectious respiratory diseases group</td>
<td>Active TB</td>
<td>1.85 ± 1.44</td>
<td>3.25</td>
</tr>
<tr>
<td></td>
<td>Non-infectious respiratory diseases group</td>
<td>0.59 ± 0.44</td>
<td></td>
</tr>
<tr>
<td>TB group vs. non-TB infections group</td>
<td>TB group</td>
<td>1.76 ± 1.27</td>
<td>3.46</td>
</tr>
<tr>
<td></td>
<td>Non-TB infections group</td>
<td>0.57 ± 0.51</td>
<td></td>
</tr>
<tr>
<td>TB group vs. noninfectious group</td>
<td>TB group</td>
<td>1.76 ± 1.27</td>
<td>3.46</td>
</tr>
<tr>
<td></td>
<td>Noninfectious group</td>
<td>0.59 ± 0.44</td>
<td></td>
</tr>
<tr>
<td>TB group vs. control group</td>
<td>TB group</td>
<td>1.76 ± 1.27</td>
<td>1.73</td>
</tr>
<tr>
<td></td>
<td>Control group</td>
<td>1.13 ± 0.45</td>
<td></td>
</tr>
</tbody>
</table>

* Significant.

Table 4
Comparison of MBL level in µg/ml inside active TB (group I), respiratory infection other than TB (group IIIa) and controls (group IV).

<table>
<thead>
<tr>
<th>Radiological features among active TB cases</th>
<th>Minimal</th>
<th>Moderate</th>
<th>Advanced</th>
</tr>
</thead>
<tbody>
<tr>
<td>Number of cases</td>
<td>5</td>
<td>24</td>
<td>1</td>
</tr>
<tr>
<td>MBL (mean ± SD)</td>
<td>1.45 ± 0.60</td>
<td>1.84 ± 1.52</td>
<td>4.00 ± 0.00</td>
</tr>
<tr>
<td>SE</td>
<td>0.27</td>
<td>0.31</td>
<td>−</td>
</tr>
<tr>
<td>Significant Test</td>
<td>0.55</td>
<td>1.38</td>
<td>3.84</td>
</tr>
<tr>
<td>P value</td>
<td>0.584*</td>
<td>0.180*</td>
<td>0.018*</td>
</tr>
</tbody>
</table>

* Difference between minimal and moderate.
** Difference between moderate and advanced.
*** Difference between minimal and advanced.

![Fig. 3. Comparison of mannose binding-lectin (MBL) (µg/ml) levels between males and females in all studied groups as well as totally.](image1)

![Fig. 4. Mean MBL distribution among the studied groups.](image2)

![Table 3](image3)

![Table 4](image4)

![Table 5](image5)

![Table 6](image6)
This study showed that MBL in about 70% of healthy control group ranges from normal to low (Table 3), this means that normal subjects have normal or low levels in the majority but not too high or too low as in patients. Some studies have found a partial protective effect of heterozygosity for MBL variant alleles (low level of MBL) against TB [12]. Also, Chen et al., (2015) [15] found that HYB haplotype could lead to decreased serum MBL and enhanced susceptibility to TB. Although same authors in a large study on Tanzanian patients (2015), found no association between MBL2 alleles, MBL levels, and tuberculosis susceptibility, other studies found that high level of MBL could reduce tubercle bacilli infections [16].

This work showed that MBL levels were higher in active than in inactive TB cases but the difference was not significant (Table 3, Fig. 2). Beside the above results this means that disease activity is associated with increased MBL levels. Denholm et al., (2010) [13] compared patients with active tuberculosis and those with a past history of infection, and found serum MBL levels to be higher in the acute phase, although this difference was small and not statistically significant. On the other hand, they had evaluated the relation between MBL2 genotype and MBL levels in a meta-analysis of, 7 studies addressing this issue and concluded that MBL2 gene is not associated with TB susceptibility, but high MBL levels could be. This result might be explained by the fact that MBL is a part of the innate immunity which is not affected by chemotherapy taken in treatment of TB so its level could remain constant before & after treatment. To our knowledge, no study has compared serial MBL levels in patients during and after active tuberculosis infection. The discrepancies between the above studies might be due to variations in numbers of cases or races that result in genetic variations.

In this study, relation of MBL to extent of disease in X-ray showed significant difference between MBL in a patient with extensive disease than both minimal and moderate disease patients while no significant difference was seen between minimal and moderate disease cases. Again, higher levels related to disease extensions could be a protective event or just a reflection to the inflammatory response. Being only one patient has extensive disease statistical comparison is not so conclusive.

In the present study ESR and temperature were high in all infectious groups especially in active TB cases and this is expected being only one patient has extensive disease statistic comparison is not so conclusive. From the above results, it could be concluded that MBL mostly play a role in increased susceptibility to tuberculosis but didn't differentiate between active and inactive disease and its role in other respiratory diseases is less clear.

References


