Risk Factors for Haematological Malignancies

Immune-mediated Diseases, Body Mass Index and Magnetic Fields

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ABSTRACT

Haematological malignancies constitute approximately 7% of all cancer cases in Sweden, and leukaemia is in fact the most common malignancy among children. The few established risk factors, such as ionising radiation and immunodeficiency, only explain a small number of the cases. The overall aim of this thesis was to examine the association between some potential risk factors and the development of haematological malignancies. Risk factors investigated were diseases affecting the immune system, excess body weight and exposure to magnetic fields.

We investigated the risk of childhood leukaemia following exposure from magnetic fields in infant incubators in a nationwide case-control study. In Sweden, 752 cases occurred among all live-born infants between 1973 and 1989, followed until the end of 1989. Magnetic field exposure was thoroughly assessed using data from medical records on length of infant incubator treatment, as well as measurements of magnetic field levels inside each incubator model. We found no association between magnetic field exposure and childhood leukaemia overall. However, elevated risk estimates were found in subgroup analyses of acute lymphoblastic leukaemia in the 5-9 year age group. Random variation is a likely explanation for the latter findings. An alternative interpretation is that a relatively long induction period is required.

We examined the influence of allergic conditions and autoimmune diseases on risk of haematological malignancies in two studies based on different materials. First, we conducted a cohort study including 16,539 Swedish twins with prospectively collected data on allergic conditions from questionnaires. Haematological malignancies registered between 1969 and 1999 were identified by cross-linkage to the Swedish Cancer Registry. A possible increase in risk of leukaemia was suggested in relation to hives, while eczema during childhood was associated with a tendency towards an elevated risk of non-Hodgkin’s lymphoma (NHL). The findings are, however, based on a small number of cases. Second, in a nationwide registry-based case-control study comprising 39,908 cases of haematological malignancies that occurred between 1987 and 1999, and 149,344 controls, occurrence of asthma and autoimmune disease was assessed from hospital discharge diagnoses. Asthma did not affect the risk of developing haematological malignancies; if anything, the results in the registry-based study indicated decreased risks. We found positive associations between Th1-mediated diseases where chronic inflammation is part of the pathogenesis, such as psoriasis and Sjögren’s syndrome, and haematological malignancies. Our findings suggest that chronic autoimmunity and immune stimulation contribute to the risk of developing haematological malignancies.

The relationship between overweight, obesity and haematological malignancies was studied in a prospective cohort study based on Swedish and Finnish twin cohorts. In total 70,194 persons were followed in the National Cancer Registries for about three decades. Our results indicate that obesity may be involved in the pathogenesis of myeloma, and excess weight possibly in the pathogenesis of chronic myeloid leukaemia and Hodgkin’s lymphoma. Risks of developing chronic lymphocytic leukaemia and NHL were not affected by excess weight.
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1 LIST OF ABBREVIATIONS

AIHA  Autoimmune haemolytic anaemia
ALL   Acute lymphoblastic leukaemia
AML   Acute myeloid leukaemia
APC   Antigen presenting cell
BMI   Body mass index
CI    Confidence interval
CLL   Chronic lymphocytic leukaemia
CML   Chronic myeloid leukaemia
EBV   Epstein-Barr virus
ELF   Extremely low frequency
HL    Hodgkin’s lymphoma
IARC  The International Agency for Research on Cancer
IBD   Inflammatory bowel disease
Ig    Immunoglobulin
IGF   Insulin-like growth factor
IGFBP Insulin-like growth factor binding protein
ITP   Idiopathic thrombocytopenic purpura
MGUS  Monoclonal gammopathy of undetermined significance
MS    Multiple sclerosis
NHL   Non-Hodgkin’s lymphoma
OR    Odds ratio
RA    Rheumatoid arthritis
RR    Relative risk
SES   Socioeconomic status
SLE   Systemic lupus erythematosus
T     Tesla
Th1   T helper lymphocyte type 1
Th2   T helper lymphocyte type 2
WHO   The World Health Organisation
2 AIMS OF THE THESIS

The overall objective of this thesis was to examine the association between diseases affecting the immune system, excess body weight, the influence of the environmental exposure to magnetic fields, and the risk of developing haematological malignancies.

The specific aims were:

✓ To assess whether exposure to magnetic fields in infant incubators increases the risk of childhood leukaemia.

✓ To examine the influence of allergic conditions on the risk of leukaemia, lymphoma and myeloma.

✓ To investigate the association between chronic immune cell activation in several Th1- and Th2-mediated autoimmune diseases, asthma and the risk of leukaemia, lymphoma and myeloma.

✓ To investigate if overweight and obesity are associated with an increased risk of leukaemia, lymphoma and myeloma.
3 INTRODUCTION

Cancer is a major threat to population health and the second leading cause of death in Sweden. In 2004, 50,384 new cases of total cancer and 3269 cases of haematological malignancies occurred\(^1\). Carcinogenesis is a process of several steps, which begins with a transformation and leads to uncontrolled clonal cell growth. Genetic alterations are necessary, but not sufficient for malignant progression.

3.1 HAEMATOLOGICAL MALIGNANCIES

Approximately 7% of all cancer cases are haematological malignancies, among both men and women\(^1\). Haematological malignancies arise from the haematopoietic or lymphoid tissue. Lymphocytic leukaemia, lymphoma and myeloma are developed from lymphocytes, whereas myeloid leukaemia stems from myeloid cells. Further subtyping is based on cell type, the stage of maturation and the level of differentiation. This can be ascertained by cell phenotyping. Acute lymphoblastic leukaemia (ALL) stems from immature lymphocytes, chronic lymphocytic leukaemia (CLL) and Hodgkin’s lymphoma (HL) from lymphocytes transformed at later stages of development, while myeloma cells are derived from the final stage of maturation of B lymphocytes, i.e. the plasma cells\(^2\).

In haematological malignancies, normal bone marrow haematopoiesis is often disrupted due to infiltration of clonal tumour cell growth. This is the main reason for the common laboratory findings of anaemia, granulocytopenia and thrombocytopenia. However, for some haematological malignancies, like myeloproliferative disorders such as polycythaemia vera and essential thrombocythaemia, erythrocyte and platelet counts may be increased. Blood leukocyte count is often elevated due to clonal expansion of malignant cells. These changes in blood cell composition are reflected in the clinical symptoms of fatigue, a tendency to bleed and bruise easily, as well as an increased susceptibility to infections. Other common symptoms are malaise, lymph node enlargement, fever, weight loss and night sweats.

Table 3.1 Number of cases and incidence of haematological malignancies per 100,000 inhabitants in Sweden 2004.

<table>
<thead>
<tr>
<th>Diagnosis (ICD-7)</th>
<th>No. of cases</th>
<th>Incidence</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Males</td>
<td>Females</td>
</tr>
<tr>
<td>Leukaemia (204-207)</td>
<td>626</td>
<td>453</td>
</tr>
<tr>
<td>ALL (2040)</td>
<td>66</td>
<td>51</td>
</tr>
<tr>
<td>CLL (2041)</td>
<td>287</td>
<td>171</td>
</tr>
<tr>
<td>AML (2050)</td>
<td>138</td>
<td>115</td>
</tr>
<tr>
<td>CML (2051)</td>
<td>43</td>
<td>48</td>
</tr>
<tr>
<td>Lymphoma (200-202)</td>
<td>906</td>
<td>725</td>
</tr>
<tr>
<td>NHL (200+202)</td>
<td>808</td>
<td>652</td>
</tr>
<tr>
<td>HL (201)</td>
<td>98</td>
<td>73</td>
</tr>
<tr>
<td>Myeloma (203)</td>
<td>305</td>
<td>254</td>
</tr>
</tbody>
</table>
Table 3.1 shows the incidence and number of cases per year of haematological malignancies in Sweden\(^1\). During the last few decades, the incidence of leukaemia has been fairly constant\(^1\). The incidence of non-Hodgkin’s lymphoma (NHL) was on the increase some decades ago, but this trend has levelled off (Figure 3.1, adapted from the National Board of Health and Welfare\(^1\)). For HL the incidence has decreased (Figure 3.2\(^1\)). The latter finding is probably due to change in diagnostic practices\(^3\). However, the fact that some cases previously diagnosed as HL are now probably classified as NHL only explains part of the elevated number of incident NHL cases. There was a slight increase in the incidence of myeloma between 1960 and 1980, but since then it has been fairly stable. The increase could be due to changes in diagnostic practices as myeloma is often diagnosed incidentally in the course of evaluating older individuals for other conditions.

**Figure 3.1 Incidence of non-Hodgkin’s lymphoma since 1960.**

![Incidence of non-Hodgkin’s lymphoma since 1960.](image1)

**Figure 3.2 Incidence of Hodgkin’s lymphoma since 1960.**

![Incidence of Hodgkin’s lymphoma since 1960.](image2)
Leukaemia is the most common type of malignancy among children and accounts for approximately one third of all cases of childhood cancer. ALL constitutes about 75-80% of the leukaemia cases, with a peak in incidence at 2-4 years of age, followed by a subsequent decline until about the age of 40; after this it increases with increasing age. A pan-European study of almost 30,000 cases of leukaemia in children and adolescents between 1978 and 1997 revealed a 0.8% yearly increase in the incidence of ALL.

In Sweden, acute myeloid leukaemia (AML) accounted for 10% of childhood leukaemia in 2004. The incidence of AML has a small peak in children under 5 years of age, followed by a slight decline until the age of 10; then it rises continuously, increasing in slope at age 50 and levels off among the elderly. Incidence rates are somewhat higher for men than for women at age 55 and older.

CLL is derived from B-lymphocytes. This is the most common type of leukaemia, constituting almost 45% of new leukaemia cases in Sweden in 2004. CLL primarily affects the elderly, and incidence is higher among men than women.

Chronic myeloid leukaemia (CML) originates from haematopoietic progenitor cells. The Philadelphia chromosome t(9:22)(q34;q11) is a diagnostic requirement for CML. Few cases occur during childhood, but the incidence increases with increasing age.

NHL is the most common type of haematological malignancy and is the 9th most common type of cancer in Sweden, constituting 3% of all cancer. NHL is the general term describing all lymphoma that are not HL. Not surprisingly, it is a heterogeneous group of malignancies which, according to the World Health Organisation (WHO) Classification of Tumours of Haematopoietic and Lymphoid Tissues in 2001, includes more than 20 subtypes of lymphoma. NHL mainly affects the elderly.

HL is named after Thomas Hodgkin, who first described the condition in 1832. The incidence of HL has a bimodal age distribution, with most cases occurring at 15-30 years of age and in persons older than 60 years.

As mentioned above, myeloma results from the clonal proliferation of malignant plasma cells. This malignant clone is capable of producing immunoglobulin (Ig), and monoclonal Ig is usually present in serum or urine of myeloma patients. In 2004, 17% of the new haematological malignancies in Sweden were due to myeloma. The incidence increases with increasing age.

### 3.2 Risk Factors

A small proportion (up to 5%) of acute leukaemia cases are associated with genetic syndromes, such as Down’s syndrome. Ionising radiation increases the risk of developing ALL. The age distribution of ALL incidence suggests that some factors are of special importance during the pre- and postnatal periods for the development of juvenile ALL, while other factors are involved in the pathogenesis of adult ALL.

In 2002, Greaves suggested the ‘two hit’ model, implying that chromosome translocations during foetal development are often the initiating events in leukaemia,
and that additional postnatal events are required for development of the disease\textsuperscript{11}. For every child diagnosed with ALL, there are at least 20 ‘healthy’ children who have had a chromosomal translocation, leading to the development of a covert preleukaemic clone generated in utero\textsuperscript{11}. Therefore the real bottleneck in development of ALL seems to be a stringent requirement for a second ‘hit’ after birth, i.e. exposure and additional chromosomal or molecular abnormality.

Infectious agents appear to be of importance in some forms of ALL, such as human T-cell lymphotrophic virus type 1 (HTLV-1) in adult T-cell leukaemia, and Epstein-Barr virus (EBV) in post-transplantation lymphoma\textsuperscript{12} and mature B-cell ALL\textsuperscript{13}. Furthermore, it has been postulated that an abnormal response to common infections plays a decisive role in the development of childhood leukaemia\textsuperscript{9,14}. Mechanisms could be infections occurring in previously unexposed or susceptible individuals due to population mixing\textsuperscript{14}, or a lack of exposure to infection and a failure of immune system modulation in infancy, followed by an abnormal immune response after delayed exposure to infectious agents\textsuperscript{9}. Ionising radiation\textsuperscript{10}, exposure to benzene and chemotherapeutic agents have all been implicated as risk factors for AML\textsuperscript{15}. Several bone-marrow-damaging pharmacological substances have also been suggested as potentially important factors in the pathogenesis of some cases of AML, e.g. chloramphenicol and methoxypsoralen. Other chemical products (e.g. pesticides and herbicides) have also been implicated, as well as cigarette smoking. However, for the large majority of cases no explanation for the leukaemic transformation can be found.

Even less is known about risk factors for chronic leukaemias. Large doses of ionising radiation is the only established risk factor for CML\textsuperscript{10,16} and, even though large differences in CLL incidence exist between geographic regions and ethnic groups, suggesting genetic or environmental influence, no clear-cut risk factors for CLL, apart from old age\textsuperscript{17}, have been identified thus far. Familial aggregation has been observed, but the nature of a possible hereditary component remains to be elucidated\textsuperscript{17}.

Most cases of NHL are sporadic and no apparent cause can be detected. However, there is an increased risk of NHL in inherited and acquired immunodeficiencies (e.g. AIDS and post-transplant immunosuppression)\textsuperscript{18}. Also, some autoimmune disorders have been identified as risk factors for NHL (e.g. coeliac disease and Hashimoto’s thyroiditis)\textsuperscript{19}. Infectious agents have also been implicated in the pathogenesis of NHL, e.g. \textit{Helicobacter pylori} in gastric NHL and EBV in Burkitt’s lymphoma\textsuperscript{19}.

HL has been reported to cluster geographically and within families, which indicates that genetic factors could be of importance in HL development. Another interpretation is that infectious agents are involved in the pathogenesis of HL. Indeed, EBV has been suggested as a risk factor in connection with HL\textsuperscript{20}. However, most cases are sporadic and other known carcinogenic factors seem to be of little significance (e.g. smoking, chemical agents and ionising radiation). The bimodal age distribution is interesting in this context, as it suggests that HL is a complex group of conditions, with different risk factors being of importance during different periods of life.

It has been difficult to establish risk factors for myeloma. Reported findings include slightly increased risks in connection with exposure to ionising radiation\textsuperscript{21}. 
occupational exposure to petroleum products and farm work. Increased risks have been observed for first-degree relatives of myeloma patients, indicating genetic predisposition as a possible risk factor. A previous diagnosis of monoclonal gammopathy of undetermined significance (MGUS), is a consistent risk factor for myeloma. However, MGUS should be considered a precursor condition that may develop into myeloma, rather than a different diagnostic entity.

In summary, very little is known about risk factors for haematological malignancies.

### 3.2.1 Magnetic Fields

We are exposed to magnetic fields from ubiquitous sources. First of all, there are static magnetic fields: primarily the static magnetic field which surrounds the earth and affects us all (in the range 25 μT-65 μT) but also, although much less common, static magnetic fields that occur e.g. in industries and in diagnostic practices in medicine, such as magnetic resonance imaging. The few animal studies that have been carried out on static magnetic fields and the haematopoietic system or cancer are equivocal, and risks of haematological malignancies in epidemiological studies are inconsistent. In addition to this, there is electric power generation, whose transmission and use result in extremely low frequency (ELF) magnetic fields. In Sweden, the frequency used for power transmission is 50 Hz. This is part of the non-ionising segment of the electromagnetic spectrum.

#### 3.2.1.1 Epidemiological studies

In 1979, Wertheimer and Leeper published the first study suggesting an association between exposure to ELF magnetic fields and childhood cancer. Exposure to magnetic fields, in particular residential magnetic field exposure from power lines and risk of childhood leukaemia, has been extensively studied since then. There was a lively debate as to whether the risk of childhood leukaemia was increased in children exposed to ELF magnetic fields. Exposure assessment has most commonly focused on exposure from nearby power lines, although later studies have captured fields from various sources in children’s homes. Estimation of magnetic fields has been dealt with differently: distance to the source, wire codes which consider both distance to and amount of current associated with various types of power-line configurations, calculated fields based on exposure from nearby sources and direct measurements (spot measurements, stationary measurements for 24 hours or 48 hours, personal measurements). It is uncertain whether these measures accurately reflect the biologically relevant exposure, which remains unknown. Two meta-analyses were performed in order to make the data from previous studies of childhood leukaemia more comparable and increase precision. The meta-analyses were based on measured and calculated magnetic fields from the data of twelve previous studies, respectively. It was found that there seem to be increased risks for highly exposed children, whereas low levels of exposure did not influence the risk of childhood leukaemia. This conclusion is further supported in two more recent studies. An increased risk of childhood leukaemia for children living close to power lines was found in a large study (9700 cases) from England and Wales, published in 2005.
As previously mentioned, magnetic fields derive not only from the vicinity of power lines, but also from electrical appliances at work and at home. The results from the studies investigating magnetic field exposure from appliances, e.g. electric blankets, sewing machines, hair dryers and television sets, do not provide a discernable pattern of increased risks associated with increased duration and frequency of use of appliances. The magnetic field level inside an incubator is often considerably higher than the exposure that can be measured in residential areas close to power lines; in some incubator models it is more than five times as high. In a study based on linkage of the Swedish Medical Birth Registry and the Swedish Cancer Registry, infant incubator treatment increased the risk of childhood myeloid leukaemia. However, length of treatment and measurements of magnetic field levels in infant incubators were not assessed. In 2002, the International Agency for Research on Cancer (IARC) classified ELF magnetic fields as possibly carcinogenic to humans (Group 2B), primarily based on findings from studies of risk of childhood leukaemia; however the evidence from studies on experimental animals was judged to be inadequate. In a comprehensive risk assessment report, the WHO agrees with the IARC’s classification of ELF magnetic fields as possibly carcinogenic to humans and conclude that, in general, the animal and in vitro studies do not support that the ELF magnetic fields are carcinogenic.

3.2.1.2 Biological mechanisms

Because ELF magnetic fields do not appear to initiate cancer, researchers have hypothesised that they may act as cancer promoters or progressors. It has been theorised that ELF magnetic fields may enhance damage from other sources (e.g. endogenous radicals), or that epi-genetic (non-genotoxic) interference in signal transduction may enhance cancer formation. However, in vitro and experimental studies on animals have been inconsistent and the biological mechanisms involved have not been identified.

3.2.2 Allergic Conditions and Autoimmune Diseases

Since the mid 20th century, Sweden and most other Western industrialised countries have experienced an increasing prevalence of allergic conditions such as asthma, rhinoconjunctivitis and eczema. Today, more than 20% of children suffer from some form of allergic symptom. However, reports indicate that the increasing prevalence of allergies has levelled off in countries with existing high prevalence among adolescents. At present there is no satisfactory explanation for this dramatic increase, but most prevailing theories involve different aspects of modern life, skewing immune responses towards a T helper lymphocyte type 2 (Th2) pattern that favours allergic conditions (see 3.2.2.1). One of the most supported theories, ‘the hygiene hypothesis’, suggests that a clean environment in early life, with little exposure to infectious agents may be a predisposing factor for the development of atopic disease. This may result in insufficient development of the T helper lymphocyte type 1 (Th1) response and would leave room for inappropriate Th2 reactions. Today, the hygiene hypothesis has been modified and also includes exposures of bioactive compounds such as endotoxins.

Autoimmune diseases, such as rheumatoid arthritis (RA), Sjögren’s syndrome and psoriasis, are not as prevalent as allergic conditions. In Sweden, the prevalence of RA was estimated to be 0.51% in 1999. Primary Sjögren’s syndrome also has a
prevalence of about 0.5\%\[^{46}\]. In the United States, an estimated 3\% of the population suffer from some form of autoimmune disease, excluding psoriasis\[^{47}\]. The prevalence of psoriasis is about 2\%\[^{48}\].

3.2.2.1 The immune system

One function of the immune system is to protect the host from invading infectious agents such as bacteria, viruses and parasites. A reflection of this is the ability to distinguish self from non-self, a mechanism that is of importance not only in the elimination of exogenous pathogens, but also in the removal of malignant cells. If the immune system is unbalanced, allergy or autoimmunity may develop.

In responding to microbial attack, the first line of the host’s defence is the unspecific innate immune system\[^{2}\]. One of the important functions of this system is to activate the adaptive immune system. The latter system works through antigen-specific responses and, in contrast to the innate immune system, provides an extra defence in case of future re-infection, thanks to the use of long-lived memory cells. The main cellular mediators of the adaptive immune response are B lymphocytes, T lymphocytes and antigen presenting cells (APCs), such as macrophages. There are two main types of T lymphocytes: T helper cells and cytotoxic T cells. T cells are activated when antigens are presented to them by APCs. Activated T helper cells can activate other immune cells through secretion of cytokines. T helper cells, when activated, become Th1 or Th2 cells. These are characterised by secretion of two distinct sets of cytokines that antagonise each other’s effects, such as interferon-\(\gamma\) (Th1) and interleukin-4 (Th2). Another subset of T lymphocytes which are now thought to be involved in immunomodulation, are the regulatory T cells\[^{49}\]. RA, Crohn’s disease, psoriasis, Sjögren’s syndrome, pernicious anaemia and multiple sclerosis (MS) are generally regarded as diseases with Th1-dominated immune responses\[^{50}\]. The immune response is skewed towards Th2 dominance in asthma, other allergic conditions, autoimmune haemolytic anaemia (AIHA), idiopathic thrombocytopenic purpura (ITP) and rheumatic fever\[^{50}\]. It has been discussed that the Th1/Th2 balance could influence immune defence and carcinogenesis. A recent review implicates a shift in immune response towards Th2 as a contributing factor in the promotion and progression of hepatocellular carcinoma\[^{51}\].

The dominant function of B cells is to produce antigen-specific Ig. The T cells are important for activation of B cells and regulation of their Ig production, including isotype switching such as to IgG or IgE\(^2\). Allergens appear to lack the capacity to induce mature germinal centres in lymph follicles. Germinal centres normally remove most B cells destined for IgE production through apoptosis. Most allergens only trigger a weak antigenic stimulation, which does not induce maturation of germinal centres and may thus lead to the development of long-lived IgE-producing plasma cells\[^{52}\].

3.2.2.2 Epidemiological studies

During the last few years, there has been an increased interest in a possible association between immune-mediated diseases and risk of developing cancer, an aspect which has been examined in a number of studies.
Studies investigating associations between allergies and non-haematological malignancies generally lack consistency or provide insufficient evidence to draw conclusions\(^5\). Despite conflicting results, strong inverse associations have been reported between allergy and pancreatic cancer and glioma, whereas asthma has been positively associated with risk of lung cancer\(^5\). A recent review of the association between allergic conditions and cancer includes published studies on haematological malignancies during the period 1966-2005\(^5\). The results concerning an association between allergy and haematological malignancies are conflicting\(^5\)\(^3\)\(^-\)\(^5\)\(^7\). No convincing evidence supports an association between a history of allergy and HL\(^5\). Furthermore, the majority of the studies on myeloma show no association with allergy. Notably, the larger cohort studies have found inverse\(^5\)\(^6\),\(^5\)\(^8\)-\(^6\)\(^1\) or no associations\(^6\)\(^1\),\(^6\)\(^2\) between asthma and haematological malignancies.

There are studies that have shown altered incidence of cancer in association with some autoimmune diseases. Studies consistently support an increased risk of lymphoma in patients with RA, whereas it seems as if the risk of malignancies in the lower gastrointestinal tract may be decreased\(^6\). Recent data support an increased risk of certain cancers in systemic lupus erythematosus (SLE) patients, and the risk appears to be most elevated for lymphoma\(^6\). The increased risk of colon cancer in persons with ulcerative colitis is well established, while it has been harder to establish whether and how much the incidence of colon cancer is increased in individuals with Crohn’s disease\(^6\). There does not appear to be an increase in the total risk of cancer outside of the gastrointestinal tract in persons with inflammatory bowel disease (IBD). Whether or not the risk of developing lymphoma in connection with IBD is increased is unclear, since studies show contradictory results. If an association between haematological malignancies and IBD exists, the impact of that association appears to be limited\(^6\).

The immune response is skewed towards Th1 in psoriasis, Sjögren’s syndrome and MS. The inflammatory pattern differs considerably between the diseases, however. In psoriasis and Sjögren’s syndrome, the inflammation is chronic and often abundant, while in MS it is intermittent and sparse. Not surprisingly, influence on cancer incidence varies between the conditions.

Most previous studies on psoriasis include very few cases. The larger studies have observed increased risk estimates for haematological malignancies\(^6\)\(^6\)-\(^7\)\(^1\), but other studies have not shown an association\(^2\)\(^4\),\(^5\)\(^6\),\(^6\)\(^7\)-\(^7\)\(^6\). However, psoriasis does not appear to increase the risk of developing myeloma\(^2\)\(^4\),\(^7\)\(^1\),\(^7\)\(^6\). A review from 2006 and a recent meta-analysis both conclude that current knowledge supports an increased risk of NHL in persons with Sjögren’s syndrome\(^6\)\(^3\),\(^7\)\(^7\). The few previous studies that have investigated the risk of NHL among persons with MS found an elevated risk estimate\(^7\)\(^8\) or risks close to the null value\(^7\)\(^9\),\(^8\)\(^0\).

Apart from asthma and other allergic conditions, AIHA and ITP also have a Th2-dominated immune response. AIHA and ITP often present as paramalignant phenomena. Their occurrence is most established in connection with CLL\(^8\)\(^1\), but they are also associated with other lymphoproliferative disorders, such as NHL\(^8\)\(^2\).
In conclusion, previous studies of the association between allergic conditions and cancer show inconsistent results, while there appears to be an association between some autoimmune disorders and risk of malignancy. However, even the latter results are not consistent between all studies.

3.2.2.3 Biological mechanisms

Two contradictory hypotheses have been proposed in order to explain the relationship between immune-mediated conditions and cancers. The immune surveillance hypothesis postulates that allergic conditions protect against development of malignancies because the immune system is enhanced and its capability to detect and destroy malignant cells is improved. As previously mentioned, there are studies that lend support to the immune surveillance hypothesis, with findings of decreased risks in association with allergy, but the results are inconsistent.

The antigenic stimulation hypothesis implies that immune-stimulating conditions increase the risk of malignancies through chronic stimulation of cells resulting in the occurrence of random mutations in actively dividing cells. If the antigenic stimulation hypothesis is correct, allergic conditions and autoimmune diseases would be expected to increase risks of haematological malignancies. As discussed above, several studies suggest a positive association between autoimmunity and haematological malignancies, although other studies have not observed an association.

The idea that inflammation and cancer are connected is not new; already in 1863, Virchow postulated that cancer originated from sites of chronic inflammation. It is not hard to appreciate the fact that inflammation is generally found in the vicinity of malignant tumours, since inflammation is part of the host’s reaction pattern in order to eliminate cancer cells. Recent data has expanded this view and it is now believed that inflammation may be important for the development of some tumours. In agreement with the antigenic stimulation hypothesis, many cancers originate in areas with chronic inflammation, such as development of colon cancer in patients with ulcerative colitis. Infectious aetiologies have been identified for some inflammation-associated malignancies; *Helicobacter pylori* may lead to gastric cancer and lymphoma, and hepatitis C virus can cause hepatocellular cancer.

3.2.3 BMI

Obesity is a major contributor to the global burden of disease. From a public health perspective, the worldwide increasing prevalence of overweight and obesity during the last few decades is a cause for concern. Approximately 15-25% of men and 10-35% of women are obese in Europe and the USA. Excess weight is a problem also among young people. The prevalence of obesity among 18-year-old men in Sweden has increased 1.7 times between 1971 and 1993. Obesity increases the risk of many diseases such as cardiovascular disease, type-2 diabetes and several different malignancies, e.g. cancers of the colon, kidney, endometrium, as well as breast cancer among postmenopausal women.

Most body fat is deposited in the subcutaneous and intraabdominal adipose tissue, in the form of triglyceride. Because fat is diffusely distributed in the body, it is
impossible to measure total body fat directly. Dual-energy X-ray absorptiometry is presently the golden standard for estimating body fat. This method can be used to validate other methods of measuring body fat, but is primarily used in research. Other methods used to estimate body fat are body circumferences, waist/hip ratio, skin fold thickness and bioimpedance (the resistance to a weak current is measured, which depends on the proportion of lean mass to fat mass).

Body mass index (BMI; weight/height$^2$; kg/m$^2$) is considered to be a good indicator of body fat in adult white individuals$^{92}$. BMI is commonly used in clinical practice as well as in large epidemiological studies. It is easy to use, as all that is needed for calculation is information on height and weight. The underlying assumption is that most of the variation in weight for persons of the same height is due to fat mass. However, BMI is limited by its inability to distinguish fat mass from muscle mass. Therefore, it is probably a less valid measure in individuals with a large amount of muscle mass and in elderly persons with diminished muscle mass. The WHO’s criteria for normal BMI is 18.5-24.9; for thinness BMI should be less than 18.5; for overweight it is in the range 25.0-29.9; and for obesity BMI is at least 30.0$^{86}$.

3.2.3.1 Epidemiological studies

As previously mentioned, obesity is known to increase the risk of developing several malignancies. However, several earlier studies have found inconsistent results when examining if there is an association between obesity and haematological malignancies$^{93-115}$. One difficulty in interpreting the results is that most studies include relatively few cases with excess weight. To our knowledge, no studies have provided evidence of a protective influence of obesity on the risk of developing haematological malignancies.

Studies regarding an association between obesity and risk of developing leukaemia have shown divergent findings. Most studies have examined risk of leukaemia overall, and in the majority of these studies increased risks have been found, both in cohort studies$^{101, 102}$ and case-control studies$^{100, 106}$. However, one cohort study found a risk increase for men but not for women$^{99}$, another had an elevated risk estimate with confidence intervals that included the null value$^{94}$. There are also cohort studies where no clear association was found$^{97, 108}$.

Several case-control studies, including one of the largest studies$^{100}$, have found elevated risks of NHL in association with excess weight$^{113-115}$. However, some studies did not find an association$^{104, 105, 112}$. Among the cohort studies, some have observed risk increases$^{99, 108}$, other studies no association$^{94, 95, 98, 102, 111}$ and a few studies different results for men and women$^{97, 110}$. Data is sparse on obesity and risk of HL, as only a few studies have addressed this potential relationship. An elevated risk was shown among men, but not women, in one study$^{97}$. In most studies no clear association has been found$^{102, 105, 111}$.

Also for myeloma, there are inconsistent findings in previous literature. Risk increases for myeloma have been observed in some studies$^{96, 99, 100}$, but not in others$^{97, 102, 108}$; and in some studies the results are inconclusive$^{93, 103}$.
3.2.3.2 Biological mechanisms

It is well-known that oestrogens play important roles in the aetiologies of both breast cancer and endometrial cancer. As oestrogens are produced in adipose tissue, hyperoestrogenaemia secondary to obesity is a possible explanation for the increased risk of breast and endometrial cancer observed in obese women. In fact, adipose tissue is the major source of oestrogen in postmenopausal women. The fact that obesity influences levels of sex-steroid hormones is further supported by findings in a recent cross-sectional study, where positive correlations were found between BMI and oestrogen in postmenopausal women, and between BMI and free testosterone in all women.

It has been proposed that an increased risk of developing malignancy in association with obesity is mediated through the insulin-like growth factor (IGF) system. Several studies have consistently indicated that high IGF-I concentrations, or a high IGF-I/insulin-like growth factor binding protein-3 (IGFBP-3) ratio, are associated with increased risks of several malignancies. A high level of IGFBP-3 may decrease the bioavailability of IGF-I. Increasing IGF-I levels with increasing BMI have been observed in some cross-sectional studies with the maximum concentrations at a BMI of about 25-26 kg/m², and decreasing levels of IGF-I with higher BMI. Furthermore, IGF-I is important in haematopoiesis. A study on cells derived from childhood ALL found increased expression of IGF-I and other IGFs compared with controls, and it was suggested that malignant lymphoblasts promote their own growth. It has been shown that IGF-1 is able to inhibit apoptosis and stimulate cell proliferation. Hence, it could function as a leukaemogenic factor by promoting leukaemic haematopoietic cell proliferation and reducing apoptosis of cells affected by malignant transformation.

Alternative explanations for how obesity could increase the risk of cancer have been suggested; dysregulation of immune response and hyperinsulinaemia. Insulin regulates energy metabolism; it enhances the synthesis of IGF-I and decreases IGFBP levels. Hyperinsulinaemia could have an effect per se or through the IGF-I system.
4 MATERIALS AND METHODS

4.1 HAEMATOLOGICAL MALIGNANCIES

We retrieved all cases of leukaemia, NHL, HL and myeloma from the Swedish and, in paper IV, also the Finnish Cancer Registries. It is required by law to report all incident cancer cases in Sweden to the Swedish Cancer Registry, which was established in 1958. All new malignant tumours are reported by the physician who diagnosed the cancer as well as by the pathologist/cytologist. This double notification system has led to a high degree of completeness; almost 96% of all patients with a malignant tumour are reported. Since pathologists have not always been involved in the diagnostic procedure, the registry is less complete for haematological malignancies: 82% for leukaemia, 96% for lymphoma, and 84% for myeloma. Further, diagnostic practices may have changed since the start of the registries; in 1960, for example, the incidence of myeloma was lower in comparison with recent decades. This could be due to more complete registration nowadays as incidental diagnosis when the patients are examined for other conditions may occur more often. In addition, some cases previously diagnosed as HL are now likely to be classified as NHL. Malignancies identified only from death certificates are not included in the Swedish Cancer Registry. The Finnish Cancer Registry was founded in 1952, and reporting has been compulsory since 1961. Cancer cases have been reported since 1953 in a similar way as to the Swedish Cancer Registry, with the additional use of information from death certificates. In the Swedish Cancer Registry, leukaemia is coded according to the International Classification of Diseases, 8th revision (ICD-8) during the investigated period, while all other malignancies are coded according to ICD-7: leukaemia (ICD-8 204.0–207.9), ALL (ICD-8 204.0), CLL (ICD-8 204.1), AML (ICD-8 205.0), CML (ICD-8 205.1), NHL (ICD-7 200 and 202), HL (ICD-7 201) and myeloma (ICD-7 203). The corresponding national classification of the Finnish Cancer Registry was used. Approximately 99% and 91% of all new cases are morphologically verified in Sweden and Finland, respectively.

4.2 STUDY DESIGN AND SUBJECTS

4.2.1 Paper I

This was a population-based case-control study based on all live-born infants in Sweden during the period 1973-89; approximately 1,700,000 infants were identified from the Swedish Medical Birth Registry. The children were followed from birth until the end of 1989. Cancer incidence and date of death were ascertained by record linkage to the Swedish Cancer Registry and the Swedish Cause of Death Registry, respectively. A total of 752 cases with childhood leukaemia (0-16 years, ICD-8 204.0–207.9) were found. Children with Down’s syndrome were excluded, as Down’s syndrome is a major risk factor for childhood leukaemia, especially myeloid leukaemia. We identified 726 eligible cases after exclusion of children with Down’s syndrome or congenital leukaemia. One control per case was randomly selected from the study base, matched on sex, birth year and month. Via linkage of the three registries, we ensured that the chosen control was alive and had not been diagnosed with leukaemia to the date of diagnosis for the matched case. No control was diagnosed with Down’s syndrome.
4.2.2 Papers II and IV

Papers II and IV were designed as cohort studies based on registries of twins treated as population-based cohorts. Paper II is based on the older cohort of twins in the Swedish Twin Registry. In Paper IV, the study population consists of two cohorts in the Swedish Twin Registry and one from the Finnish Twin Cohort. Incidence of haematological malignancies, emigration and date of death were identified through linkage with the National Cancer Registries and Population Registries and the Swedish Cause of Death Registry.

The Swedish Twin Registry is the largest twin registry in the world. It was established in 1961 when questionnaires were sent to all 25,778 individuals in same-sexed twin pairs born between 1886 and 1925, and where both twins in the pair were living in Sweden. A response from both twins in a twin pair was required for inclusion in the Swedish Twin Registry (21,870 individuals). This older Swedish cohort was sent additional questionnaires in 1963, 1967 and 1970 (the latter to non-respondents in 1967) and the twins were further followed up as individuals, irrespective of their twin-sibling’s status.

Paper II is based on the 16,539 respondents to the 1967 questionnaire, who were still alive and had not been previously diagnosed with a haematological malignancy on 1st January 1969. The subjects were followed from this date until diagnosis of a haematological malignancy, death or the end of the study (31st December 1999), whichever came first. From the Swedish Cancer Registry we identified 134 cases of leukaemia (3 cases of ALL, 67 cases of CLL, 31 cases of AML, 7 cases of CML and 26 cases of unspecified leukemia), 112 cases of NHL, 10 cases of HL and 75 cases of myeloma. In total, 324 subjects with haematological malignancies were identified.

Paper IV includes both the older and a younger cohort in the Swedish Twin Registry. The younger Swedish cohort consists of twins born between 1926 and 1967 who were alive and living in Sweden in 1970. During 1972/73, a questionnaire was sent out to every same-sexed twin pair born from 1926 to 1958; in total 30,279 persons responded. At baseline (1st March 1974), twins that were alive and not previously diagnosed with a haematological malignancy were included.

In paper IV, we extended the study population to also include the Finnish Twin Cohort of adult same-sexed twin pairs of known zygosity born before 1958 with both co-twins alive in 1975: 25,882 persons. The twin pairs were compiled from the Central Population Registry of Finland in 1974. Questionnaires were administered to all twins in 1975, 1981 and 1990. We included all subjects 18 years or older at baseline, who were still alive and had not previously been diagnosed with a haematological malignancy at baseline (1st January 1976).

The age distribution of the Swedish twin cohorts differ considerably, with a median age at baseline of 56 years in the older cohort and 31 years in the younger cohort. To increase age comparability between the Swedish and Finnish twins, we divided the Finnish cohort into two subgroups. We combined the older Swedish and Finnish twins into an older cohort, whereas the younger twins from both countries were combined
into a separate cohort (minimum age 18 years at baseline). The cohorts were followed from baseline (1969, 1974 or 1976 depending on date of completed questionnaire) until December 2002 in Sweden and until December 2004 in Finland. In total, 740 cases of haematological malignancies (ICD-7; 200-203, ICD-8; 204-207) were identified; 282 cases of leukaemia, 291 cases of NHL, 34 cases of HL and 142 cases of myeloma. The leukaemias consist of 14 cases of ALL, 129 cases of CLL, 67 cases of AML and 22 cases of CML.

4.2.3 Paper III

In this population-based case-control study, we retrieved from the Swedish Cancer Registry all incident cases of haematological malignancies that occurred in Sweden from 1987 to 1999 – in total 39,908 cases. We identified 12,397 cases of leukaemia, 2394 cases of HL, 18,186 cases of NHL and 7189 cases of myeloma. Only haematological malignancies that occurred at least one year after diagnosis of exposure, i.e. diagnosis of autoimmune disease or asthma from the Swedish Hospital Discharge Registry, were included. Continuously throughout the study period, two controls per cancer case were randomly selected from the Swedish Population Registry, stratified on sex, five-year age groups, and year of diagnosis. For the youngest cases (< 20 years), five controls for each cancer case were retrieved in order to increase the precision. Controls were selected simultaneously for parallel studies of pancreatic cancer and brain tumours, and the entire set of controls was used in the analyses – in total 149,344 controls.

4.3 EXPOSURE ASSESSMENT

4.3.1 Paper I - Magnetic Fields

Information about pre- and neonatal conditions and treatments, including incubator treatment, was extracted blindly with regard to case-control status from medical records at each hospital. For each child we determined whether the child had been treated in an infant incubator, and if so, the duration of the treatment. We also determined if any of the hospitals routinely treated children in infant incubators. We collected information about which infant incubator models had been used at each hospital, and during which periods of time these incubators were used.

**Figure 5.1 Measurement of magnetic fields in an infant incubator.**
Magnetic field levels inside different types of incubators were determined through measurements of the magnetic fields. In collaboration with a physicist and after interviewing an experienced nurse at a neonatal ward, a standard procedure for measurement of the magnetic fields inside incubators was established. It resembled the actual nursing conditions of the treated infant as closely as possible. We measured each incubator at three defined spots corresponding to the position of the infant’s body, for 15 minutes at each spot, in at least three different incubators of each model when possible. All measurements in each incubator were summarised using the arithmetic mean and the median. For each model, we calculated the mean value and the average of the medians based on the measured incubators of the same model.

The exposure of each child was assessed by combining the duration of the treatment in an infant incubator with the magnetic field values of the incubator models used at the hospital at the time of treatment. The medical records did not contain information about which incubator model the infant had been treated in. If more than one model was available at the hospital when the infant was treated, the magnetic field level for that infant was assessed as the average of the magnetic field levels in the available incubators. In the main analyses we only used the information on incubator use extracted from the medical records. However, we also performed analyses where we included routine treatment in the exposure assessment.

Several different measures of magnetic fields in infant incubators were used:

- Treatment versus no treatment
- Exposure time (unexposed, ≤30 hours, >30 hours)
- Individual magnetic field values, dependent on incubator model and based on arithmetic mean (unexposed, ≤0.6 µT, >0.6µT)
- Cumulative exposure; length of treatment combined with the arithmetic mean of the measured magnetic field value (unexposed, ≤10 µT-hours, >10 µT-hours)

4.3.2 Papers II and IV - Allergic Conditions and BMI

Self-reported information was prospectively collected in the questionnaires sent out to the twins in the Swedish and Finnish twin cohorts.

4.3.2.1 Allergic conditions

In Paper II, information about allergic conditions from the Swedish 1967 questionnaire was used. Both present and past asthma, hay fever, eczema and hives were asked for (yes/no). A questionnaire was also sent to the older Swedish twins in 1963, where the subjects could mark if they had had asthma or eczema. These answers were used if the answers from the corresponding questions in 1967 were missing.

4.3.2.2 Body Mass Index

In Paper IV, BMI was used as a measure of relative body weight and was calculated from self-reported current weight and height in the questionnaire. Assessment of BMI is based primarily on the questionnaires administered in 1967 for the older Swedish cohort, in 1972/73 for the younger Swedish cohort and in 1975 for the Finnish cohort. We analysed BMI categorised according to the WHO’s criteria; BMI<18.5 (thinness), BMI 18.5-24.9 (normal, reference category), BMI 25.0-29.9 (overweight), BMI ≥30.0
(obesity) and as a continuous variable. Persons with extreme values of BMI, height or weight were excluded from analysis, as these were likely to be data errors (BMI<14, BMI>50 if improbable weight in comparison with height, height<140 or >220 cm or weight <40 or >200; in total 108 observations). After all exclusions, there were 25 179 individuals in the older cohort and 45 015 individuals in the younger cohort (when the Swedish and Finnish cohorts were combined). BMI was higher in the older cohort than in the younger cohort.

4.3.3 Paper III - Autoimmune Diseases and Asthma

In 1964, the Swedish National Board of Health and Welfare started to collect data on individual hospital discharges in the Swedish Hospital Discharge Registry. The coverage of the Swedish population has gradually increased and encompassed 60% of the population in 1969, 85% by the end of 1983 and 100% in 1987. For all cases and controls we extracted information about all hospital discharges with a diagnosis of 36 different autoimmune diseases or asthma from 1969 to 1999. The discharge diagnoses consist of one principle diagnosis and up to five contributory diagnoses until 1996; since 1997, up to 7 contributory diagnoses. The diagnoses are coded according to ICD-7 (1964-1968), ICD-8 (1969-1986), ICD-9 (1987-1996) and ICD-10 (1997-). On the most detailed level, approximately 83% of the diagnostic codes are correct for primary diagnoses. We investigated asthma and common autoimmune diseases that required hospital inpatient care: psoriasis, Sjögren’s syndrome, pernicious anaemia, MS, AIHA, ITP and rheumatic fever. For Sjögren’s syndrome, we performed additional analyses where subjects with a diagnosis of RA or SLE were excluded. The registration is based on individual discharges rather than on individuals. In the analyses we used the first date of registration with a specific diagnosis if the disease was registered on more than one occasion in the Hospital Discharge Registry.

4.4 CONFOUNDERS AND EFFECT MODIFIERS

In Paper I, we adjusted for potential confounders from reproductive history (earlier pregnancies, spontaneous abortions), during pregnancy (assisted reproduction, twin pregnancy, hypertension, smoking, infection, diagnostic X-rays and ultrasound), maternal age at the newborn infant’s birth and caesarean section. Further, we adjusted for possible risk factors related to the newborn infant (firstborn child, gestational age, birth weight, malformations, use of supplementary oxygen, jaundice, infections, diagnostic X-rays, postnatal asphyxia and treatment at a neonatal ward).

In Paper II, adjustments for age at enrolment and sex were performed for all analyses. Further, we controlled for potential confounding from alcohol consumption (g/month), level of education, and smoking habits (non-smokers, former smokers, current smokers).

In Paper III, all analyses were adjusted for sex, age at diagnosis (cases) or age at selection (controls) and socioeconomic status (SES). Information about SES was obtained from the censuses of 1985 and 1990. For controls and malignancies diagnosed during the period 1987-1990 the categories were based on occupational status from the census of 1985, while for controls and malignancies diagnosed after 1990 they were based on the census of 1990. Statistics Sweden’s has categorised SES from 1985 and
1990 into 13 categories. We constructed a socioeconomic variable with five categories based on this categorisation; blue-collar workers, lower and intermediate white-collar workers, senior white-collar workers and self-employed (excluding farmers), farmers and a fifth category of unclassifiable employees and those where there was a lack of information on profession.

In Paper IV, we adjusted for age at enrolment, sex and country in all analyses. In separate analyses, we have controlled for potential confounding from alcohol consumption (reference category up to the 3rd quartile at 213 g/month, with the upper quartile divided into 2 categories at 720 g/month), level of education (elementary school or less, education above elementary school), physical activity at work (sedentary/active/physically strenuous), recreational physical activity (physically inactive, intermediate activity, hard physical training), diabetes mellitus (yes/no according to self-report) and smoking habits (non-smoker, former smoker, current smoker).

4.5 STATISTICAL METHODS

4.5.1 Papers I and III

Odds ratios (OR) and their 95% confidence intervals (CI) were used as measures of association. The OR can be considered as an estimate of the incidence rate ratio in case-control studies; given that certain study design and sampling criteria have been fulfilled. We adjusted for relevant covariates described above. In Paper I, conditional logistic regression was used to investigate the association between magnetic field exposure in infant incubators and childhood leukaemia in this 1:1 matched case-control study. ALL and AML were analysed both together and separately. For ALL, we also stratified for age at diagnosis (0-4, 5-9, 10-16 years). In Paper III, unconditional logistic regression was performed to study the association between asthma, autoimmune diseases and haematological malignancies. In all analyses, we controlled for sex, SES and age at diagnosis (cases) or age at selection (controls). In the main analyses, induction periods of 1 year were used, i.e. we only included haematological malignancies diagnosed at least 1 year after hospital discharge diagnosis of autoimmune disease or asthma. Additional analyses with induction periods of 5 and 10 years were also performed.

4.5.2 Papers II and IV

For each haematological malignancy in the Swedish and Finnish twin cohorts, we calculated the relative risks (RR) with 95% CIs using Cox’s Proportional Hazards Model. Potential confounders were adjusted for in separate analyses. To ensure that CIs were not erroneous due to dependencies within twin pairs we performed analyses that adjusted variance estimates for correlated outcomes. We accomplished this through the use of a SAS macro, which results in adjusted CIs being generally more conservative than unadjusted ones, as the twins are generally positively correlated for traits. If correlations within twin pairs do not differ from what is observed between unrelated individuals in the cohort with respect to cancer risk, adjusted and unadjusted variance estimates are identical. Relative risk estimates are not altered by this procedure.
5 RESULTS

5.1 PAPER I - MAGNETIC FIELDS AND CHILDHOOD LEUKAEMIA

We studied the effect of different aspects of magnetic fields and the risk of childhood leukaemia. There were no differences in risk for children treated in infant incubators compared with those who were untreated for all leukaemia combined (OR= 1.0, 95% CI 0.7-1.4), ALL (OR=0.9, 95% CI 0.6-1.3) or AML (OR=1.8, 95% CI 0.5-6.0). Risks in association with more than 30 hours of treatment were not affected for leukaemia overall (OR=1.2, 95% CI 0.6-2.1), for ALL (OR=1.1, 95% CI 0.5-2.1) or for AML (OR=0.7, 95% CI 0.1-4.0). However, the risk estimate was elevated for AML among those exposed for a maximum of 30 hours, albeit with a wide CI (OR=4.0, 95% CI 0.4-35.8), but not for leukaemia overall or ALL.

The individual magnetic field levels varied 15-fold between the infants. Analyses based on individual magnetic field values showed risks close to unity for leukaemia overall (OR=0.9, 95% CI 0.5-1.5) or ALL (OR=0.7, 95% CI 0.4-1.2) for children in the lower exposure category (with a maximum exposure of 0.6 μT), while for AML the OR was 4.0 (95% CI 0.4-35.8). No increased risks were found for leukaemia overall (OR=0.9, 95% CI 0.5-1.7), for ALL (OR=0.9, 95% CI 0.5-1.7) or for AML (OR=1.0, 95% CI 0.1-7.1) in the highest exposure category (>0.6 μT). Similar results were obtained for analyses using a cut-off point of 1.0 μT. In age-stratified analyses of ALL (0-4, 5-9, 10-16 years), the risk estimates were elevated for children aged 5-9 years: OR 2.4 (95% CI 0.6-10.5) for the lower exposure category and OR 5.0 (95% CI 0.5-47.5) among infants exposed to 0.6 μT or more. These risk estimates were, however, based on small numbers of exposed cases (6 and 5 exposed cases, respectively).

Combining length of treatment with the arithmetic mean of the measured magnetic field value to estimate the cumulative exposure in μT-hours, showed no associations between treatment up to 10 μT-hours and all leukaemia combined (OR=0.6, 95% CI 0.3-1.3) or ALL (OR=0.4, 95% CI 0.2-1.1). There were no exposed controls for AML. Children treated in infant incubators for more than 10 μT-hours had risk estimates close to the null value for leukaemia overall (OR=1.0, 95% CI 0.6-1.6), ALL (OR= 1.0, 95% CI 0.6-1.6) and AML (OR= 0.8, 95% CI 0.2-3.4).

5.2 PAPER II - ALLERGIC CONDITIONS

We evaluated the influence of different allergic conditions on the risk of developing haematological malignancies and found elevated risks or risks close to the null value. Tendencies towards increased risks of leukaemia in association with asthma (RR=1.6, 95% CI 0.8-3.5) and hives (RR=2.1, 95% CI 1.0-4.5) were observed. The positive association between hives and leukaemia was further elevated when CLL was excluded (RR= 3.6, 95% CI 1.6-8.5). Eczema during childhood tended to increase the risk of NHL (RR=2.3, 95% CI 1.0-5.3). All risk estimates were decreased for the different allergic conditions in association with myeloma (range of RRs 0.5-0.9), but the CIs included unity. We achieved similar results when we performed additional analyses with induction periods of 5 years (Figure 5.1).
5.3 PAPER III - AUTOIMMUNE DISEASES AND ASTHMA

In Paper III, we observed several positive associations between autoimmune diseases and leukaemia, CLL, AML, CML and NHL. With the exception of asthma, the numbers of exposed cases with ALL and HL were too few for meaningful separate analyses. Generally, the ORs for myeloma were close to unity.

We found an increased risk of leukaemia, excluding CLL, (OR=1.6, 95% CI 1.1-2.3) for persons with an earlier diagnosis of psoriasis. Risk estimates in association with psoriasis remained elevated, although the CIs included the null value, after induction periods of 5 years (OR=1.5, 95% CI 1.0-2.4) and 10 years (OR=1.4, 95% CI 0.8-2.5). Psoriasis was associated with increased risks of NHL (OR=1.6, 95% CI 1.3-2.1) in the main analysis, as well as in our additional analyses with induction periods of 5 years (OR=1.6, 95% CI 1.2-2.1) and 10 years (OR=1.6, 95% CI 1.1-2.2).

In order to identify primary Sjögren’s syndrome, we performed analyses where we excluded subjects with RA or SLE. Primary Sjögren’s syndrome was positively associated with NHL (OR=7.5, 95% CI 3.5-15.9), also with induction periods of 5 years (OR=8.1, 95% CI 3.4-19.5) and 10 years (OR=6.0, 95% CI 2.1-17.5). The risk estimates were elevated for the other haematological malignancies in separate analyses, but with wide CIs. By combining all haematological malignancies we could increase the number of exposed cases, and precision. We found an increased risk of all haematological malignancies combined for primary Sjögren’s syndrome (OR=4.0, 95% CI 1.9-8.2); after 5-year and 10-year induction periods the risks were 3.7 (95% CI 1.5-9.0) and 2.8 (95% 1.0-8.0).
Analyses of an association between MS and NHL suggested a reduced risk, OR=0.5 (95% CI 0.3-1.0), also after induction periods of 5 and 10 years OR=0.5 (95% CI 0.2-1.0).

We observed an elevated risk of leukaemia (OR=6.5, 95% CI 3.3-12.8) in association with AIHA. Risks were also increased in separate analyses of AIHA and leukaemia, excluding CLL, (OR=7.0, 95% CI 3.1-16), CLL (OR=6.7, 95% CI 2.7-17) and AML (OR=8.0, 95% CI 3.0-21.0). Further, there was a positive association between AIHA and NHL (OR=12.0, 95% CI 7.1-20.2). Increased risks were found for leukaemia (OR=4.3 95% CI 2.3-7.8) and NHL (OR=3.4, 95% CI 1.9-6.2) following a diagnosis of ITP. The risk estimates were elevated in subtype analyses of leukaemia: leukaemia, excluding CLL, (OR=5.4, 95% CI 2.8-10.5), CLL (OR=2.5, 95% CI 0.8-8.1), AML (OR=6.6, 95% CI 2.8-15.5) and CML (OR=6.6, 95% CI 1.6-27.3). In general, induction periods of 5 and 10 years resulted in less elevated, but still increased, risk estimates for the different types of haematological malignancies in association with AIHA or, to a lesser extent, ITP.

The risks of haematological malignancies tended to be decreased or not affected in association with asthma. Analyses of asthma showed reduced ORs for leukaemia, leukaemia other than CLL, CLL, AML, NHL and all haematological malignancies combined, whereas risks of CML and myeloma were equal to unity (Tables 5.1-5.3). The ORs of the different haematological malignancies following a diagnosis of asthma after induction periods of 5 and 10 years were reduced or not influenced (Tables 5.1-5.3).

**Table 5.1 Asthma and risk of haematological malignancies with induction periods of 1 year, 5 years and 10 years.**

<table>
<thead>
<tr>
<th>Induction period</th>
<th>All haematological malignancies</th>
<th>Leukaemia (204-207)</th>
<th>Leukaemia (excl. CLL)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>No.</td>
<td>OR</td>
<td>95% CI</td>
</tr>
<tr>
<td>1 year</td>
<td>561</td>
<td>0.9</td>
<td>0.8-1.0</td>
</tr>
<tr>
<td>5 years</td>
<td>352</td>
<td>0.8</td>
<td>0.7-0.9</td>
</tr>
<tr>
<td>10 years</td>
<td>211</td>
<td>0.9</td>
<td>0.7-1.0</td>
</tr>
</tbody>
</table>

**Table 5.2 Asthma and risk of leukaemia with induction periods of 1 year, 5 years and 10 years.**

<table>
<thead>
<tr>
<th>Induction period</th>
<th>ALL (2040)</th>
<th>CLL (2041)</th>
<th>AML (2050)</th>
<th>CML (2051)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>No.</td>
<td>OR</td>
<td>95% CI</td>
<td>No.</td>
</tr>
<tr>
<td>1 year</td>
<td>10</td>
<td>0.6</td>
<td>0.3-1.0</td>
<td>70</td>
</tr>
<tr>
<td>5 years</td>
<td>2</td>
<td>0.2</td>
<td>0.1-0.8</td>
<td>48</td>
</tr>
<tr>
<td>10 years</td>
<td>2</td>
<td>0.5</td>
<td>0.1-1.9</td>
<td>27</td>
</tr>
</tbody>
</table>

**Table 5.3 Asthma and risk of lymphoma and myeloma with induction periods of 1 year, 5 years and 10 years.**

<table>
<thead>
<tr>
<th>Induction period</th>
<th>NHL (200 + 202)</th>
<th>Hodgkin’s lymphoma (201)</th>
<th>Myeloma (203)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>No.</td>
<td>OR</td>
<td>95% CI</td>
</tr>
<tr>
<td>1 year</td>
<td>254</td>
<td>0.9</td>
<td>0.8-1.1</td>
</tr>
<tr>
<td>5 years</td>
<td>164</td>
<td>0.8</td>
<td>0.7-1.0</td>
</tr>
<tr>
<td>10 years</td>
<td>101</td>
<td>0.9</td>
<td>0.7-1.1</td>
</tr>
</tbody>
</table>
5.4 PAPER IV - BMI

At baseline, the median age of the older cohort was 55 years and the corresponding value for the younger cohort was 30 years. The results for the combined older and younger twin cohorts are presented in Table 5.4. We found that excess body weight tended to be associated with CML, HL, myeloma and, to some extent, ALL. We observed an indication of an increased risk for all haematological malignancies combined in association with BMI as a continuous variable and with obesity for the older twins (RR=1.4, 95% CI 1.0-2.0).

BMI did not influence the risks of all leukaemia combined or, in subtype analyses, of CLL in either cohort. Overweight tended to increase the risk of ALL both in the older cohort (RR=2.8, 95% CI 0.2-32.8) and in the younger cohort (RR=3.5, 95% CI 0.9-12.9); for continuous BMI we found a RR of 1.12 (95% CI 0.99-1.28) and a RR of 1.18 (95% CI 1.07-1.31), respectively. We observed a tendency towards increased risk of AML in association with overweight among women (RR=2.1, 95% CI 1.0-4.4) in the older cohort, but not among men (RR=1.2, 95% CI 0.4-3.5). Overweight was associated with an elevated risk of CML when the two cohorts were combined, as well as in separate analyses of the older (RR=2.6, 95% CI 0.9-7.8) and younger cohort (RR=4.1, 95% CI 0.8-20.6).

In our material, the risk of NHL was not affected by overweight or obesity. The risk estimate for HL was increased twofold in association with overweight when the older and younger cohorts were combined. Seven cases of HL occurred in the younger cohort (RR=2.7, 95% CI 1.0-7.0), six of them among Finnish twins (RR=6.5, 95% CI 1.8-24.2) and one case in the younger Swedish twin cohort (RR=0.6, 95% CI 0.1-4.0).

We observed an increased risk of myeloma in association with obesity for the older twins (RR=2.7, 95% CI 1.5-4.9), as well as a trend of increasing risk of myeloma with increasing BMI (RR=1.08, 95% CI 1.03-1.14 per unit increase).
Table 5.4 Age- sex- and country-adjusted relative risks (RR) and 95% confidence intervals (CI) for haematological malignancies in relation to body mass index (BMI) at baseline for all 70,194 Swedish and Finnish twins.

<table>
<thead>
<tr>
<th>Diagnosis (ICD-7)</th>
<th>BMI</th>
<th>Nc</th>
<th>RR</th>
<th>95% CI</th>
<th>Nc</th>
<th>RR</th>
<th>95% CI</th>
<th>Nc</th>
<th>RR</th>
<th>95% CI</th>
<th>Nc</th>
<th>RR</th>
<th>95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>-18.49</td>
<td>18.50-24.99&lt;sup&gt;1&lt;/sup&gt;</td>
<td>25.00-29.99</td>
<td>30.00-</td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>All haematological malignancies</td>
<td>7</td>
<td>0.4</td>
<td>0.2-0.9</td>
<td>456</td>
<td>1.0</td>
<td>0.9-1.2</td>
<td>237</td>
<td>1.0</td>
<td>0.9-1.7</td>
<td>40</td>
<td>1.2</td>
<td>0.9-1.7</td>
<td>1.03</td>
</tr>
<tr>
<td>Leukaemia (204-207)</td>
<td>1</td>
<td>0.1</td>
<td>0.0-1.1</td>
<td>176</td>
<td>1.0</td>
<td>0.8-1.4</td>
<td>93</td>
<td>1.1</td>
<td>0.5-1.6</td>
<td>12</td>
<td>0.9</td>
<td>0.5-1.6</td>
<td>1.02</td>
</tr>
<tr>
<td>CLL (2041)</td>
<td>0</td>
<td>-</td>
<td>86</td>
<td>1.0</td>
<td>0.5-1.1</td>
<td>36</td>
<td>0.8</td>
<td>0.3-1.6</td>
<td>7</td>
<td>1.0</td>
<td>0.5-2.1</td>
<td>1.02</td>
<td>0.96-1.07</td>
</tr>
<tr>
<td>Other</td>
<td>1</td>
<td>0.2</td>
<td>0.0-1.8</td>
<td>90</td>
<td>1.0</td>
<td>1.0-2.0</td>
<td>57</td>
<td>1.4</td>
<td>0.6-2.4</td>
<td>5</td>
<td>0.8</td>
<td>0.3-2.1</td>
<td>1.03</td>
</tr>
<tr>
<td>ALL (2040)</td>
<td>0</td>
<td>-</td>
<td>8</td>
<td>1.0</td>
<td>0.8-1.5</td>
<td>6</td>
<td>2.7</td>
<td>0.9-9.5</td>
<td>0</td>
<td>-</td>
<td>-</td>
<td>1.13</td>
<td>1.02-1.26</td>
</tr>
<tr>
<td>AML (2050)</td>
<td>0</td>
<td>-</td>
<td>40</td>
<td>1.0</td>
<td>0.6-1.4</td>
<td>25</td>
<td>1.4</td>
<td>0.4-5.2</td>
<td>2</td>
<td>0.7</td>
<td>0.2-3.1</td>
<td>1.04</td>
<td>0.98-1.10</td>
</tr>
<tr>
<td>CML (2051)</td>
<td>1</td>
<td>2.9</td>
<td>0.3-27.3</td>
<td>9</td>
<td>1.0</td>
<td>1.1-6.9</td>
<td>12</td>
<td>2.8</td>
<td>0.6-13.2</td>
<td>0</td>
<td>-</td>
<td>-</td>
<td>1.04</td>
</tr>
<tr>
<td>NHL (200+202)</td>
<td>4</td>
<td>0.6</td>
<td>0.2-1.6</td>
<td>186</td>
<td>1.0</td>
<td>0.7-1.2</td>
<td>87</td>
<td>1.0</td>
<td>0.6-1.5</td>
<td>14</td>
<td>1.1</td>
<td>0.6-1.9</td>
<td>1.02</td>
</tr>
<tr>
<td>HL (201)</td>
<td>2</td>
<td>1.9</td>
<td>0.4-8.8</td>
<td>19</td>
<td>1.0</td>
<td>1.0-4.3</td>
<td>13</td>
<td>2.0</td>
<td>0.7-15.0</td>
<td>0</td>
<td>-</td>
<td>-</td>
<td>0.99</td>
</tr>
<tr>
<td>Myeloma (203)</td>
<td>0</td>
<td>-</td>
<td>82</td>
<td>1.0</td>
<td>0.7-1.5</td>
<td>46</td>
<td>1.0</td>
<td>0.7-1.5</td>
<td>14</td>
<td>2.2</td>
<td>1.2-3.9</td>
<td>1.07</td>
<td>1.02-1.12</td>
</tr>
</tbody>
</table>

N<sub>c</sub> = No. of cases. <sup>1</sup>Reference category.
6 DISCUSSION

6.1 MAIN RESULTS

6.1.1 Magnetic Fields

Most previous studies on magnetic fields and risk of childhood leukaemia have examined residential magnetic field exposure. Two meta-analyses, where several previous studies were pooled, found no influence on risk of childhood leukaemia at low levels of exposure, whereas the risk was doubled in the higher exposure category\textsuperscript{28, 29}. To our knowledge, Paper I is the only study conducted on exposure to magnetic fields in infant incubators and risk of childhood leukaemia. Our intention was to find a new approach to estimate magnetic field exposure through a new study design in an environment with high magnetic field levels, i.e. infant incubators.

An increased risk of childhood myeloid leukaemia in association with infant incubator treatment was shown in a previous register-based study where several prenatal and neonatal risk factors were evaluated\textsuperscript{38}. These risk factors were examined for childhood lymphatic leukaemia using the same material, and no indication of risk increases was found in association with infant incubator treatment\textsuperscript{127}. Our study is an extension of this material. The objective was to investigate whether the previously noted risk increase of childhood myeloid leukaemia in association with infant incubator treatment was due to magnetic field exposure and also to examine its influence on risk of childhood lymphatic leukaemia with more complete assessment of magnetic field exposure. We could assess magnetic field exposure more thoroughly by abstracting data from medical records including information on length of infant incubator treatment for the study participants, retrieving information about which specific infant incubator models were in use at the hospitals when the children were born and by measuring the magnetic field levels inside the different infant incubator models. The lack of association between infant incubator treatment and lymphatic leukaemia is in accordance with our main findings. In a subgroup analysis of ALL in children aged 5-9 years we found increased risk estimates, which could be due to the necessity of an induction period of this length or, considering the wide confidence intervals, perhaps more likely, to chance. However, an elevated risk would be in agreement with Greaves ‘two-hit’ model\textsuperscript{11}, where magnetic field exposure could be the ‘second hit’.

In separate analyses of AML, we found elevated risk estimates in the lower exposure category both for length of incubator treatment as well as for level of magnetic field exposure, which is in accordance with the earlier register-based study on childhood myeloid leukaemia\textsuperscript{38}. However, risk was not influenced in the highest exposure category. Further, the results could be explained by random variation due to few exposed cases.

Magnetic field exposure in infant incubators differs in several aspects from magnetic field exposure examined in earlier studies. Most previous studies have investigated residential exposure, or conducted measurements on individuals. In either case, these exposure assessments are estimates of longer periods of magnetic field exposure. In
contrast, length of treatment in infant incubators is much shorter, often less than 24 hours. On the other hand, magnetic field levels are generally higher in infant incubators than in homes close to power lines\textsuperscript{37}. Another difference is that infant incubator treatment occurs at the same age for all children, whereas it may take place at any time for other exposure sources. An advantage of our study compared with studies on magnetic fields from electrical appliances is that recall bias is not a problem\textsuperscript{33-36}. Our findings provide little evidence for an increased risk of childhood leukaemia overall following high levels of magnetic field exposure during a short period early in life.

6.1.2 Allergic Conditions and Autoimmune Diseases

In accordance with our findings, most larger previous studies have observed positive associations between psoriasis and NHL\textsuperscript{66}, lymphoma\textsuperscript{68, 69} and non-lymphocytic leukaemia\textsuperscript{70, 71}, respectively. In contrast, no risk increases have been shown in other studies for NHL\textsuperscript{56, 71-74}, leukaemia\textsuperscript{73, 75} or AML\textsuperscript{67}. Similar to our result, the few previous studies on myeloma have not shown increased risks\textsuperscript{24, 71, 76}. Our study included a considerably larger number of persons with haematological malignancies and psoriasis than most previous studies. One large study on AML found a similar risk estimate with precision similar to that of our study\textsuperscript{67}. Two large studies of psoriasis and NHL showed a lack of association\textsuperscript{56, 72}. In both studies, analyses were based on self-reported psoriasis and therefore it is probable that more persons with less severe psoriasis are included compared with studies based on hospital-diagnosed psoriasis. An explanation for the differing results could be that the elevated risk of developing malignancy is limited to severe psoriasis. Support for this explanation was found in a study where patients with severe psoriasis had a highly increased risk of developing a lymphoproliferative malignancy, while persons with less severe psoriasis only had a twofold increase in risk\textsuperscript{132}. However, these findings must be interpreted with caution, as there are some methodological considerations. For instance, the unexposed group consisted of patients with hypertension and therefore it is uncertain whether they are representative of the study population. In a cohort study from 2006, a slightly increased risk of NHL was found in association with mild psoriasis, while severe psoriasis did not influence the risk\textsuperscript{69}. The latter risk estimate was based on only four cases and is therefore inconclusive. If there is a higher risk for lymphoproliferative malignancies in patients affected by severe psoriasis, it remains to be established whether this is attributable to psoriasis per se or to the treatments given. Severe psoriasis involves systemic treatment with potent drugs such as cyclosporine and methotrexate, both of which are well known to increase the risk of malignancy. Indeed, in a recent study of a cohort of psoriasis patients treated with psoralen-UV-A an increased risk of lymphoma was found following treatment with methotrexate, but otherwise the risk was comparable with that in the general population\textsuperscript{69}.

Previous studies are consistent concerning a risk increase for NHL in patients with Sjögren’s syndrome; our results are in agreement with this\textsuperscript{63, 77}. Our findings of elevated risks of haematological malignancies are largely attributable to NHL, though the risk estimates were increased for haematological malignancies in separate analyses. To our knowledge, our study is the first to have demonstrated increased risks of haematological malignancies and NHL, after an induction period of 10 years following a diagnosis of Sjögren’s syndrome.
There was no positive association between MS and haematological malignancies in our data. Instead, we found an indication of a protective effect of MS on risk of NHL. Previous data is sparse for risk of NHL in patients with MS; the risk estimate was increased in one study and risks were close to unity in two other studies.

Our findings of remaining increased risk estimates following diagnoses of, mainly, AIHA but also ITP with induction periods of at least 5 and 10 years before haematological malignancy was diagnosed could indicate that AIHA and ITP may not solely be paramalignant phenomena. Instead, they may precede the development of haematological malignancy; alternatively, the malignancy has been present as long as 10 years before diagnosis. In a material of 107 patients diagnosed with AIHA, with no known malignancy, 17 developed CLL or lymphoma within 9-76 months. In either case, this is an important consideration in the surveillance of AIHA and ITP patients to enable early diagnosis and treatment of haematological malignancies. However, as the analyses are based on few exposed cases more studies are warranted to confirm these results.

Although it has been commonly assumed that the allergic immune response offers little benefit to the individual, it has been argued that it may offer a valuable evolutionary advantage in terms of protection from parasitic infections.

The literature is inconsistent regarding an association between conditions of allergic disease and haematological malignancies. One explanation could be that risks may differ according to the type of allergic disease (asthma, allergic rhinitis or eczema) with or without association with development of allergic sensitisation (presence of IgE antibodies to common allergens). Further, most previous studies are retrospective case-control studies, where recall bias and selection bias might have been present. However, decreased or not affected risk estimates have mostly been found for haematological malignancies in association with asthma. The larger cohort studies found no association or reduced risks of haematological malignancies in association with asthma. Two of them, however, were based on cancer mortality and not incidence, which makes them harder to interpret because survival also affects the outcome.

This is partly in contrast to our results in Paper II, where asthma and other allergic conditions either did not influence or tended to increase the risks of haematological malignancies. In Paper III, however, asthma did not affect or tended to reduce the risks of haematological malignancies. Introduction of induction periods of 10 years following a diagnosis of asthma emphasised the decrease in risk of leukaemia (excluding CLL), especially for AML. These findings lend some support to the immune surveillance hypothesis (an improved capability to eliminate malignant cells). In a previous study no association between asthma and AML was found.

Asthma, other allergic conditions and autoimmune diseases appear not to influence the risk of myeloma in our data, which is in accordance with the majority of previous studies. In Paper III, we observed tendencies towards reduced risks in association with asthma (Th2-mediated) following induction periods of 10 years, in
particular of leukaemia (excluding CLL) and AML. These findings support the immune surveillance hypothesis.

We observed that Th1-mediated diseases where chronic inflammation is part of the pathogenesis increase the risk of haematological malignancies. Conditions that alter the normal function of the immune system may not only result in an impaired ability to suppress activation of autoimmune cells, but also lead to dysfunctional elimination of malignant haematological cells. An interesting finding in support of the importance of chronic inflammation in the development of haematological malignancies is the finding in a large recent study that risk of lymphoma in patients with RA was associated with severity of disease; not with the treatments\(^{134}\). In conclusion, the antigenic stimulation hypothesis is supported for Th1-mediated diseases in combination with chronic inflammatory processes.

### 6.1.3 BMI

The literature is inconsistent regarding an association between excess weight and haematological malignancies. In our study, we found no effect of obesity on the risk of developing CLL. Previous epidemiological research on the association of obesity with CLL is sparse, and results inconsistent; a few studies have indicated elevated risks\(^{102, 106}\). However, a case-control study did not find an association\(^{105}\), one cohort study of older women found no risk increase\(^{101}\), and another cohort study found an increased risk of B-CLL only in a secondary analysis of the effect of BMI at 50 years of age\(^{98}\). CML was positively associated with overweight in our study, which has been described for myeloid leukaemia also in a recent cohort study\(^{107}\), while a large cohort study based on hospital discharge, diagnosis of obesity did not observe an association\(^{102}\). In an earlier, large case-control study there was a tendency towards a risk increase of overweight, whereas obesity elevated the risk of CML\(^{106}\).

We found no association between BMI and NHL. Our findings are in accordance with several cohort studies\(^{94, 98, 111}\), including the largest cohort study (hospital discharge diagnosis of obesity)\(^{102}\), which found no association between obesity and NHL. Another cohort study found no association either, but it is possible that some cases were not registered as follow-up of incident cases was based on self-report\(^{95}\). However, in some cohort studies elevated risks have been shown, e.g. for women with a BMI \(\geq 35\) and for men in a large study, which however was based on cancer mortality, and therefore could reflect the combined influence of BMI on cancer incidence and on survival\(^{99}\). Further, an elevated risk estimate for NHL was found in a study on Korean men with a BMI \(\geq 27\) or more\(^{108}\). Two other studies observed a risk increase for NHL in association with obesity in women, but not in men\(^{97, 110}\). Some case-control studies have shown increased risks\(^{109, 113-115}\), including one of the largest studies\(^{100}\), while others have not found an association\(^{104, 105, 112}\). In summary, the studies regarding excess body weight and risk of NHL are inconsistent.

Only a few studies have examined the relationship between obesity and HL. Our results are in agreement with an earlier cohort study where a risk increase was observed, but was restricted to men\(^{97}\). Further, in two recent cohort studies\(^{102, 111}\), and one case-control study\(^{105}\), overweight and obesity did not affect the risk of HL.
There was an elevated risk of myeloma among obese persons in our older cohort, which is in accordance with several earlier studies\textsuperscript{96, 99, 100}. In two of these studies separate analyses were performed for both men and women, in which increased risk estimates were found for both sexes. In contrast, some cohort studies have not found a clear association\textsuperscript{97, 102, 108}. In a cohort study on postmenopausal women, the risk estimate was elevated but the CI included the null value\textsuperscript{103}. The risk of myeloma was elevated among both men and women in our study, whereas in an earlier cohort study increased risk estimates were observed for men but not women\textsuperscript{93}. Thus, studies to date are not consistent.

It is well known that obesity may elevate the risk of some malignancies such as colon cancer, breast cancer and endometrial cancer\textsuperscript{86, 89, 90}. Our findings suggest that excess weight may also contribute to development of myeloma and, possibly, CML and HL, whereas we found no evidence for influence on risks of CLL and NHL. Different pathogenesis between subtypes of leukaemia, NHL, HL and myeloma could explain these findings. However, random variation cannot be ruled out as an explanation. Further research in larger epidemiological studies is needed to clarify a potential relationship between obesity and haematological malignancies.

6.2 METHODOLOGICAL CONSIDERATIONS

6.2.1 Validity

Generalisability, or external validity, refers to how inferences can be drawn from results in a study to risk in a more general population. In the included studies in this thesis, generalisability appears to be a minor problem, with the possible exception of Paper III where hospitalised patients may constitute a selection of individuals with more severe disease. This would affect generalisability of our results, as they may primarily be applicable to people with severe disease. The Swedish Twin Registry is considered a study base that is representative of the general population of Sweden and has been used in many epidemiological studies\textsuperscript{135-137}.

Below, I will discuss potential bias in the context of internal validity, i.e. if we really have investigated what we intended to examine in each study. A high degree of internal validity is necessary to be able to draw conclusions.

6.2.1.1 Selection bias

Selection bias occurs when the study population is not representative of the study base of all eligible subjects; i.e. when the probability for an individual to be included in the study is related both to exposure and outcome. Selection bias may arise when persons are selected if not all cases that occur in the study base are registered or, in case-control studies, if controls are not randomly selected from the study base. There may also be selection bias if participation differs between cases and controls, e.g. the response rate when exposure is assessed from questionnaires.

We have no reason to believe that selection bias is a problem in our studies. Papers I and III are population-based case-control studies based on nationwide register data,
where all incident cases were retrieved and controls were randomly selected from the study base. In addition to register data, in Paper I we used data from medical records to assess infant incubator treatment. Because Papers II and IV are cohort studies, selection bias due to differential participation is not an issue, as the participants are asked about exposure before diagnosis of outcome and it is unlikely that non-participation is related both to exposure and outcome. However, it may occur if follow-up is different for exposed and unexposed persons. As all participants are followed up via nationwide cancer registries the risk of selection bias is probably minimal.

6.2.1.2 Exposure assessment

In Paper I, exposure information on infant incubator treatment was extracted from medical records and on magnetic field levels from measurements inside infant incubators. To avoid differential exposure misclassification we collected data from medical records blindly with regard to case-control status. Medical records were found for the majority of cases and controls. Moreover, differential exposure misclassification is unlikely as information in the medical records was noted when the child was newborn, hospitalised and, on indication, nursed in an infant incubator, which was before developing leukaemia. Non-differential exposure misclassification resulting in diluted risk estimates is likely to affect the results as we interviewed the hospital personnel about which models of infant incubators were in use 11–27 years ago. Furthermore, it was not noted in the medical records which exact model of infant incubator the child was nursed in, though several models were often in use at the ward during the same time period. However, for 59% of the children the incubator with the lowest exposure was in the same category as the incubator with the highest exposure, with the cutpoint at 1.0 µT the corresponding number was 82%. Analyses with the cutpoint at 1.0 µT did not change the results. In addition, at some hospitals all newborn children were put in infant incubators as a routine without registration in the medical records. However, separate analyses with estimated time of routine treatment included in the exposure metric did not change the results. The levels of magnetic field exposure were thoroughly assessed by measurements inside the infant incubator models that were in use at the different hospitals during the time periods the children were treated in them. This enabled investigation of a possible dose-response relationship. However, non-differential exposure misclassification could dilute the risk estimates and result in a lack of risk increases or distortion of a possible dose-response pattern.

In Papers II and IV, the study populations are cohorts with prospectively collected exposure information on allergic conditions and BMI from questionnaires, and are therefore not subject to differential exposure misclassification. This is in contrast to case-control studies assessing exposure retrospectively from self-reports, where recall bias may influence the risk, and differential misclassification of exposure occurs.

Non-differential exposure misclassification is a concern in Papers II and IV, as allergic conditions and BMI are self-reported. In general, good agreement was found between the self-reported conditions and an allergologist’s diagnosis when the validity of the allergic conditions was investigated in a group of subjects from the Swedish Twin Registry. Furthermore, some support for the postulated hypothesis that allergic conditions are inversely associated with glioma risk was demonstrated in a study using
the same data on allergic conditions and occurrence of brain tumours. Self-reported weight and height are generally accurate and do not contribute materially to errors in assessing BMI. However, non-differential misclassification and attenuated effect estimates can be a problem among overweight and obese persons when BMI is self-reported, because of their tendency to underestimate weight. The study participants are followed from baseline for about three decades, depending on questionnaire date. This long time period makes it plausible that non-differential misclassification of exposure occurs if persons who are unexposed at baseline become exposed, and vice versa. However, classification of having had childhood eczema is not influenced because the youngest individuals in our cohort were 42 years old when the questionnaire was sent out. Hay fever and allergic asthma usually present at a younger age, which implies that such bias would be of minor importance. Changes in BMI could have diluted our effect estimates towards the null value. However, our cohorts consist of an ageing population as they are closed. If most people gain weight with increasing age, some truly exposed individuals are classified as unexposed, resulting in diluted risk estimates for analyses of overweight and obesity. When we analysed the effect of excess weight on risk of haematological malignancies, we divided the material into an older and a younger cohort because changes in BMI (and non-differential misclassification) probably have a greater impact among younger persons due to assessment of BMI at a much younger age and a longer period of follow-up compared with the older subjects. This approach made it possible to examine both the effect of BMI at younger ages on the risk of developing haematological malignancies later in life, as well as the influence of BMI at older ages on the disease risk. Non-differential exposure misclassification is probably less important among older persons, as BMI is estimated closer to time of diagnosis, and probably closer to the BMI during the years preceding cancer diagnosis, than in the younger cohort.

In Paper III, a case-control study design using nationwide register data was used to assess exposure of asthma and autoimmune diseases. Generally, case-control studies based on information from registries may be biased by differential registration of exposure in cases compared with controls. We gathered exposure information prior to diagnosis from the Swedish Hospital Discharge Registry. Nevertheless, there is a risk of differential misclassification of exposure if cases (or controls) are more likely to become classified with asthma or autoimmune disease. In our study, this would happen if hospital discharge diagnoses of asthma and autoimmune diseases occur more often (or seldom) among the cases. We have been able to reduce possible differential misclassification of exposure by examining whether longer induction periods between diagnoses of autoimmune disease or asthma and incident malignancy (up to 10 years) influence the results, compared with shorter induction periods. Our findings that elevated risk estimates remained after induction periods of 5 years and 10 years following a diagnosis of psoriasis for leukaemia (excluding CLL) and NHL strengthen the assumption that our results are not biased by differential classification of exposure. Non-differential misclassification of exposure could affect our results. It is a strength that asthma and autoimmune diseases are diagnosed by a physician. Validations of the Hospital Discharge Registry by analysing medical records have demonstrated a correct ICD-code at the five-digit-level in 83% of all primary diagnoses. It is important to remember that people with asthma and autoimmune diseases who see a general practitioner but are not hospitalised, are not included. The patients in our study are
likely to suffer from more severe disease than outpatients, e.g. patients with asthma and psoriasis.

6.2.1.3 Outcome classification

The outcome, i.e. haematological malignancy, was assessed from the Swedish Cancer Registry for all studies in this thesis. For Paper IV, we extended the study population to include the Finnish Twin Cohort and data on outcome was retrieved from the Finnish Cancer Registry as well. The Finnish Cancer Registry uses similar procedures for registration as the Swedish Cancer Registry.

In Paper I, differential misclassification of outcome is unlikely, as we find no reason to believe that information about infant incubator treatment would affect diagnosis of leukaemia. Similarly, Papers II and IV are unlikely to be affected by differential outcome misclassification, as we do not believe that it is likely that a diagnosis of haematological malignancy among twins in Sweden and Finland was affected by having an allergic condition or excess weight. In Paper III, subjects with immune-mediated diseases could have been subject to increased clinical surveillance, resulting in differential diagnosis of malignancies compared with the general Swedish population. We dealt with this potential problem by requiring an induction period of at least one year after diagnosis of immune-mediated disease for inclusion of cases with haematological malignancy. An argument against bias due to differential misclassification of the outcome is that the obtained results were in accordance with analyses where induction periods of at least 5 and 10 years were used. In addition, risk estimates for asthma were not elevated which would be expected if screening bias of outcome had an impact on the risks.

Non-differential misclassification of malignancy, resulting in diluted risk estimates, is probably of minor importance because two independent notifications systems ensure high coverage and high completeness of the Swedish Cancer Registry. Physicians and pathologists are required by law to report all new cases of cancer to this registry. However, non-differential misclassification of outcome may be a problem in analyses of specific subtypes of haematological malignancies, as diagnostic practices may have changed during follow-up. In particular, some cases previously diagnosed as HL are now likely to be classified as NHL.

6.2.1.4 Confounding

In all our studies, we have adjusted for several potential confounding factors that did not affect the results materially. Age and sex were adjusted for in every analysis. In spite of control from potential confounding, we cannot rule out that residual confounding may have affected the results to some extent. In Paper I, we performed separate analyses where we controlled for diagnostic X-rays for children who were nursed in infant incubators. In Papers II-IV, concerning allergic conditions, autoimmune diseases and BMI, we find it unlikely that they are related to, or that the results are confounded by, ionising radiation. We controlled for smoking and alcohol in Papers II and IV. Unfortunately, information on smoking and alcohol habits was not available in the registries used in Paper III.
6.2.2 Precision

Random error is larger in small studies with few exposed cases. This is because there is considerable variability in the results, which are more likely to be due to chance as compared with a large study including many exposed cases. Many of our analyses are based on a small number of exposed cases resulting in wide confidence intervals, in particular for subgroup analyses. Hence, we cannot rule out chance as an explanation for most findings. For the main analyses in Paper I, a sufficient number of exposed cases were included to make it unlikely that chance is an explanation. The results on allergic conditions in Paper II and asthma in Paper III tended to contradict each other, for which there could of course be several explanations. However, in the context of precision, the larger number of exposed cases in Paper III could offer an explanation in favour of these findings for asthma.
7 GENERAL DISCUSSION AND FUTURE RESEARCH

Of all new cancer cases in Sweden every year, 1 out of 14 is a haematological malignancy. The few established risk factors for haematological malignancies explain only a small proportion of the cases. Many people are affected by allergic conditions and obesity. It would be important from a public health perspective to identify causal risk factors. In the studies included in this thesis, we investigated some potential risk factors: magnetic fields, allergic conditions, autoimmune diseases and excess weight. Further research is needed to clarify the impact of these and other potential risk factors on the development of haematological malignancies.

The pathogenesis for different subtypes of leukaemia, HL, NHL and myeloma, as well as biological mechanisms for different risk factors is likely to differ. In addition, medical treatment of allergic conditions, autoimmune diseases and obesity may affect the risk of developing cancer.

Our study of magnetic field exposure in infant incubators and risk of childhood leukaemia is unique. A large number of epidemiological studies have been conducted to examine the relationship between residential magnetic fields and childhood leukaemia. Future epidemiological studies would need to have a large number of highly exposed persons or an innovative study design to provide new insights.

Additional studies on allergic conditions, autoimmune diseases, overweight and obesity are needed to confirm and clarify our findings. The role of the immune system in cancer development requires further clarification.

Future research could include large prospective epidemiological studies on individuals with strictly defined exposures, making it possible to examine a dose-response pattern, and taking into account duration and severity of allergic conditions, autoimmune diseases and excess weight, as well as information about pharmacological treatment. This information could preferably be combined with detailed data on haematological malignancies to assess subtypes according to modern classifications including cell lineage, morphology, immunophenotype, genetic features and clinical syndromes. Furthermore, research on plausible mechanisms in animal and in vitro studies is needed in order to put the findings in epidemiological studies into biological perspective.
8 CONCLUSIONS

The main focus in this thesis was to examine the risk of developing haematological malignancies in relation to allergic conditions, autoimmune diseases, excess body weight and environmental exposure to magnetic fields. The following conclusions can be drawn:

✓ High levels of magnetic field exposure experienced over a short period of time early in life do not seem to influence the risk of developing childhood leukaemia.

✓ Asthma seems not to affect the risk of developing haematological malignancy. If anything, the findings suggest a tendency towards decreased risk of several haematological malignancies, considering in particular that long induction periods resulted in several reduced risks and tendencies to decreased risk estimates following a diagnosis of asthma.

✓ A possible increase in risk of leukaemia was suggested in relation to hives, and of NHL associated with eczema during childhood.

✓ The influence of autoimmune diseases on risk of haematological malignancies is complex. Chronic autoimmunity and immune stimulation appeared to contribute to the development of haematological malignancies. Th1-mediated diseases where chronic inflammation is part of the pathogenesis, such as psoriasis and Sjögren's syndrome, increased the risk of haematological malignancies.

✓ Increased risks of haematological malignancies following a diagnosis of AIHA tended to remain with induction periods of 5 and 10 years; for ITP a similar tendency was observed. These findings suggest that AIHA and ITP may not solely be paramalignant phenomena. However, they are based on few exposed cases, and future research is needed to clarify whether AIHA and ITP could precede the development of haematological malignancies.

✓ Risk of myeloma does not seem to be affected by allergic conditions or autoimmune diseases.

✓ Obesity appears to increase the risk of myeloma, at least among older persons. It is possible that excess weight influences the risks of CML and of HL.

✓ Risks of CLL and NHL do not seem to be influenced by overweight or obesity.
9 SAMMANFATTNING (SUMMARY IN SWEDISH)

Cancer är en folksjukdom och är den näst vanligaste dödsorsaken i Sverige. År 2004 inträffade 3 269 nya fall av hematologiska maligniteter i landet, vilket utgör ungefär 7% av alla cancerfall. Leukemi är den vanligaste typen av cancer hos barn. Joniserande strålning är en av de fåtaliga kända riskfaktorerna för hematologisk malignitet, men de förklarar endast en liten andel av de inträffade fallen. Syftet med denna avhandling var att studera om risken att utveckla hematologiska maligniteter påverkas av de potentiella riskfaktorerna allergiska och autoimmuna sjukdomar, övervikt och fetma samt exponering för magnetfält.


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