COMMITTEE ON TOXICITY OF CHEMICALS IN FOOD, CONSUMER PRODUCTS AND THE ENVIRONMENT

Third draft statement on the effects of soya consumption on thyroid status.

Introduction

1. A 2003 Committee on Toxicity (COT) report on phytoestrogens and health identified individuals with hypothyroidism as a subgroup of the population of potential concern for adverse effects of phytoestrogens in soya, and made recommendations for research. During 2014 the Committee considered the results of several FSA-funded research studies on phytoestrogens including the third arm of T05029: “The effect of soya phytoestrogen supplementation on thyroid status and cardiovascular risk markers in patients with subclinical hypothyroidism: a randomized double blind crossover study”. As a result the Committee considered that it would be timely to review evidence related to potential effects of phytoestrogens on thyroid function, which had become available since the 2003 COT report on phytoestrogens and health. The first draft statement (TOX/2014/41) summarising the new literature concerning potential risks from consumption of soya phytoestrogens on thyroid function, with particular emphasis on the risks to individuals with hypothyroidism was presented to Members in December 2014.

2. The second draft statement in Annex A has been revised taking into account previous discussion and incorporating details requested by Members in December. A table summarising effects on isoflavone exposure on thyroid function reported in human studies and further details on tabulated studies have been included. Certain sections of the document have been reworded. Additional editorial changes have also been made.

Questions on which the views of the Committee are sought

3. Members are invited to comment on the structure and text of the second draft statement.

Secretariat
March 2015

COMMITTEE ON TOXICITY OF CHEMICALS IN FOOD, CONSUMER PRODUCTS AND THE ENVIRONMENT

Second draft statement on the effects of soya consumption on thyroid status.

Background

1. A 2003 COT report on phytoestrogens and health[1] identified individuals with hypothyroidism as a subgroup of the population who might be vulnerable to adverse effects of phytoestrogens in soya, and made a number of recommendations for research. These included investigation of possible interactions of phytoestrogens with the thyroid gland in people with compromised thyroid function. The Food Standards Agency subsequently commissioned studies to address that recommendation and invited the Committee to consider the results when they became available. Having done so, the Committee concluded that it would be timely to review all evidence which had become available since 2003, concerning potential effects of phytoestrogens on thyroid function. This statement summarises the findings of that review and the COT's conclusions. The focus is principally on human studies because they were judged to be the most informative. The criteria that were employed in the literature search are set out in Annex 1.

Phytoestrogens

2. Phytoestrogens are naturally produced by some edible plants. They have been shown to affect biological processes mainly through their structural similarities to oestrogens, and their ability to bind to oestrogen receptors (ERs), thereby interfering with hormonal control mechanisms in humans and animals. The largest group of phytoestrogens are flavonoids, which comprise three subclasses – coumestans, prenylated flavonoids and isoflavones (e.g. genistein, daidzein).

The thyroid gland

3. The thyroid gland produces hormones involved in the regulation of metabolism, bodyweight and oxygen requirements, as well as growth and development during childhood. The hormones concerned are thyroxine (T_4) and triiodothyronine (T_3), which are synthesised from iodine and the amino acid tyrosine. The production of T_3 and T_4 is controlled by thyroid stimulating hormone (TSH), which is secreted by the pituitary gland and regulated by the hypothalamus.

[1] http://cot.food.gov.uk/cotreports/cotwgreports/phytoestrogensandhealthcot

4. More than 99% of circulating thyroid hormones are bound to plasma proteins, from which they can be liberated to enter cells. The thyroid hormone-binding proteins are thyroxine-binding globulin (TBG), transthyretin (TTR or thyroxine-binding prealbumin), human serum albumin (HSA) and lipoproteins. TBG has the highest affinity for T_4, and binds 75% of T_4 in serum, whilst TTR binds 20% and HSA 5%. TTR is the most important thyroid hormone-binding protein in cerebral spinal fluid (CSF).

5. Hypothyroidism is a condition that occurs when the thyroid gland is underactive and the concentrations of free (unbound) T_3 and T_4 are below the normal range. Low circulating levels of free T_3 and T_4 lead to increased secretion of TSH by the pituitary gland, and are one of the causes of goitre (enlargement of the thyroid gland).

Potential effects of phytoestrogens on thyroid function

6. In addition to their oestrogenic properties, some phytoestrogens, notably genistein and daidzein, have a similar chemical structure to thyroid hormones. It has been hypothesised that they might interact with the thyroid gland by a number of possible mechanisms, leading to interference with its normal function.

7. *In vitro* and animal studies have shown that phytoestrogens can inhibit thyroperoxidase (TPO), an enzyme involved in the synthesis of T_3 and T_4, which reduce concentrations of T_3 and T_4 in the blood and cause increased secretion of TSH (COT, 2003). The potential of genistein and diadzein to inhibit TPO has been confirmed in a high-throughput screening assay, using rat thyroid microsomes and a fluorescent peroxidase substrate Paul *et al*. (2014). Moreover, genistein and daidzein have been shown to inhibit the formation of T_4 *in vitro* in a concentration-dependent manner with IC_{50} values of approximately 2 µM (0.5 µg/mL) for genistein and 8.8 µM (2.24 µg/mL) for daidzein (Divi *et al*., 1997; BfR, 2007). However it is not clear whether such concentrations would occur *in vivo*.

8. Ebmeier and Anderson (2004) reported that genistein and daidzein can also inhibit the activity of human intrathyroidal sulphotransferases, which are involved in the inactivation and elimination of thyroid hormones.

9. Another possibility is that phytoestrogens interfere with the binding of thryroid hormones to transporter proteins. In serum and CSF, genistein and related isoflavones are highly effective inhibitors of binding by T_4 and T_3 to TTR, with a dissociation constant (Kd) of 40 nmol/L, suggesting a high affinity and equimolar binding to that of T_4 (Green *et al*., 2005; Radovic *et al*., 2006; BfR, 2007). This could affect the distribution of thyroid hormones in the body (Green *et al*., 2005; Radovic *et al*., 2006; BfR, 2007).

10. It has been hypothesised also that phytoestrogens might increase levels of thyroxine binding globulin (TBG), which could transiently increase binding capacity

for thyroxine, leading to lower levels of free T_4 and a compensatory increase in the secretion of TSH (COT, 2003).

Previous conclusions

The COT Report on Phytoestrogens and Health (2003)

11. The COT report in 2003 noted that animal studies had shown that high levels of dietary soya and isoflavones could affect thyroid function, and might have a goitrogenic effect in rodents deficient in dietary iodine. However, data from human studies suggested that isoflavones were unlikely to affect thyroid function in normal individuals with adequate iodine intake.

12. No data were found to indicate that maternal ingestion of phytoestrogens during pregnancy influences the development of the thyroid gland. However, the COT considered it possible that, together with low iodine intake, increased metabolic demands during pregnancy and increased need for thyroxine, maternal consumption of soya products could adversely influence the neurological development of the fetus.

13. A number of scientific publications evaluated by the Committee in 2003 reported cases in the 1950s and 1960s of thyroid disease (mostly goitre) associated with consumption of soya-based formula, and there was also a report of increased faecal loss of orally administered thyroxine in an athyreotic hypothyroid infant when fed soya formula as compared with cows' milk formula. This had suggested that increased faecal mass and fibre content of the diet may alter entero-hepatic circulation of thyroxine. As a consequence, changes had been made made in the processing and formulation of infant formulae (supplementation with iodine and replacement of soya flour with soya protein isolate), following which, no further reports of goitre were published.

14. The COT identified individuals with hypothyroidism as a subgroup of the population of potential concern, and the 2003 report concluded that consumption of phytoestrogen supplements, or a soya-rich diet, might provide sufficient concentrations of phytoestrogens to interfere with the T_4 replacement medication, which is given to patients with hypothyroidism. Although no adverse effects in hypothyroid children or adults had been reported in the published literature, the report noted that research had not addressed this question specifically. In view of the increasing availability of phytoestrogen-rich food and supplements in the UK, the report recommended that research be conducted on the plasma levels of T_4 in children and adults with hypothyroidism, who consume large quantities of dietary phytoestrogens.

Conclusions reached by organisations in other countries

15. A number of concerns were raised in a 2007 risk assessment by the German Federal Institute for Risk Assessment (BfR). The BfR concluded that available

toxicological studies showed that isoflavones, when consumed at high doses (for example by menopausal women), can impair the functioning of the thyroid gland. The BfR advised against long-term consumption of isoflavone-containing products made from soya (BfR, 2007).

16. A review by the American Academy of Pediatrics (AAP) Committee on Nutrition concluded that the evidence for adverse effects of dietary soya isoflavones on human development, reproduction and endocrine function was inconclusive (Bhatia *et al*., 2008). The National Toxicology Program (NTP) considered that the risk of developmental or reproductive toxicity (including thyroid effects) following consumption of soya infant formula could not be determined because data from studies in humans and experimental animals were insufficient (NTP, 2010). They identified a number of requirements for new information that would enable an improved assessment of risks.

17. Taking into account concerns that had been raised with respect to possible negative effects of soya isoflavones on sexual, reproductive and neurobehavioral development, immune function and thyroid function, the European Food Safety Authority (EFSA) recommended that concentrations of isoflavones in infant formula and follow-on formula should be kept as low as is feasible (EFSA, 2014).

Food Standards Agency-funded research

18. The research commissioned by FSA following the 2003 COT report included three projects to assess: a) the effect of soya phytoestrogen supplementation on thyroid status and cardiovascular risk markers in patients with subclinical hypothyroidism; b) the effects of soya in men with type 2 diabetes; and c) the effects of soya in women within two years of the onset of the menopause.

19. The first project comprised three independent parts (details are specified in Table 1). Each used a similar randomised crossover design, whereby a treatment (different in each part) was administered daily for two months, followed by a two-month wash-out period, and then a second alternative treatment for a further two months. The treatments in the three parts were:

- Part 1: 30 g isolated soya protein (isoflavone-free) with 16 mg of isoflavones (representative of a vegetarian diet) or 30 g isolated soya protein (isoflavone-free) with 2 mg of isoflavones (representative of a Western diet);
- Part 2: 30 g isolated soya protein (isoflavone-free) with 60 mg of isoflavones (equivalent to ingestion of phytoestrogen supplements) or 30 g isolated soya protein (isoflavone-free) alone;
- Part 3: 30 g of isolated soya protein (isoflavone-free) alone, or 30 g casein protein alone (as control).

20. Although not entirely consistent, results from the first two parts of the project both pointed to effects. The first part suggested a threefold increase in the risk of

developing overt hypothyroidism following dietary supplementation with 16 mg soya phytoestrogens in individuals with subclinical hypothyroidism (Sathyapalan *et al*., 2011). In the second part, which used a higher dose of isoflavones (60 mg), fewer patients developed overt hypothyroidism while receiving isoflavones, and although there was an increased risk, it was not statistically significant. The authors suggested that the failure to find a stronger effect with the larger dose might have been because of a higher drop-out rate and because there were fewer women in the study sample. The Committee noted that although these were possible explanations, the difference in transition to overt hypothyroidism could also have occurred by chance. The third part of the project was conducted to clarify whether effects associated with the consumption of soya protein in combination with isoflavones were attributable to the soya protein. None of the patients in the third part developed overt hypothyroidism during the study.

21. Together, the results of the three parts of the project indicate that soya protein alone does not have an effect on thyroid function in patients with sub-clinical hypothyroidism. Thus, any effects observed in the first two parts of the study appear to have depended on the exposure to isoflavones. The Committee was informed by the researchers of an earlier study in which they had investigated effects on cardiovascular risk factors in patients with diabetes, following administration of 132 mg of isoflavones alone for four weeks (Gonzalez *et al*., 2007). Although not included in their published report, data on thyroid function had been measured as a secondary end-point, and no effect on thyroid function had been observed (Atkins personal communication). They had speculated that the effect of isoflavones might depend on simultaneous exposure to soya proteins.

22. Thyroid function was also evaluated in the studies investigating the effects of soya in men with type 2 diabetes (three months treatment)[2] and women within two years of the onset of the menopause (six months treatment)[3] (Table 1). These were again randomised crossover studies, and were undertaken by the same researchers. There was no effect of soya protein alone (without isoflavones). However, when soya protein was administered together with 66 mg isoflavones, a significant increase in TSH and reduction in serum thyroxine was observed in both studies. Although levels remained within the normal range, the consistency of the changes in thyroid hormone levels that followed consumption of soya protein containing phytoestrogens, both in women within 2 years after the onset of menopause, and in men with type II diabetes and subclinical hypogonadism, supported the possibility of risks from soya ingestion in people with subclinical or overt hypothyroidism.

Other new data

23. A summary is provided below of other relevant findings that have emerged since the 2003 COT report was published. Further detail can be found in Table 1.

[2] The effect of soya protein with and without isoflavones in men with type 2 diabetes and subclinical hypogonadism – A randomized double blind parallel study. University of Hull.
[3] Soya protein with isoflavones reduce bone turnover markers in women during their early menopause – A randomised double blind placebo controlled parallel study. University of Hull.

Studies of children with thyroid dysfunction

24. Conrad *et al*. (2004) retrospectively analysed the medical records of infants diagnosed with congenital hypothyroidism and seen at a hospital during their first year of life. Two groups of patients were distinguished: a soya diet group consuming exclusively soya infant formula, who started on treatment with levothyroxine (a synthetic derivative of T_4) at a median age of 15 days (n=8), and a non-soya diet group, who started treatment at 17 days (n=70). There was no significant difference between the groups in serum levels of TSH and T_4 levels before the start of treatment with levothyroxine. However, significant differences occurred subsequently in: time to TSH normalisation (four times longer in soya group; p=0.02); first serum TSH concentration measured after treatment began (six times higher in soya group; p<0.01); and percentage of infants with increased TSH at 4 months of age, and throughout the first year of life (approximately four times higher in soya group). The authors suggested that infants on thyroxine-replacement who were fed soya needed close monitoring of free thyroxine and TSH, as they might require higher doses of levothyroxine.

25. Fruzza *et al*. (2012) described two patients with congenital hypothyroidism who, although on levothyroxine treatment, were persistently hypothyroid. The patients were girls aged 3 weeks and 5 years, and had been consuming soya-based formula and soya milk respectively (no details of isoflavone levels were available). When the consumption of soya-based products was discontinued and the dose of levothyroxine decreased, their thyroid function normalised slowly over the course of a few weeks.

Studies in adults with normal thyroid function

26. In a randomised double-blind, placebo-controlled clinical trial, post-menopausal women not on hormone replacement therapy (age 64-83 years) received a supplement (containing 90 mg of isoflavone) (n=22) or placebo (maltodextrin) (n=16) per day, plus a multi-vitamin and mineral supplement, daily for six months. Serum levels of TSH, T_4 and T_3 were measured at baseline and after 90 and 180 days. No statistically significant effects on thyroid hormone measurements were recorded in this healthy iodine-replete group of subjects (Bruce *et al.*, 2003).

27. In a randomised cross-over study, healthy young men (20-40 years old) received diets supplemented with three protein powders (milk protein isolate, and low- and high-isoflavone soya protein isolates), each for 57 days, separated by 4 week wash-out periods (Dillingham *et al.*, 2007). The low- and high-isoflavone soya protein isolates provided 1.64 ± 0.19 mg isoflavones/day (mean ± SD) and 61.7±7.4 mg isoflavones/day, respectively. Urine samples confirmed a significantly (p<0.0001) higher excretion of isoflavones and their metabolites in participants when receiving the high isoflavone diet compared to the low isoflavone diet and milk protein isolate. Blood was collected on days 1, 29 and 57 and analysed for total and free T_3 and T_4, TSH and TBG. No significant changes were recorded in any thyroid parameters when the low- and high-isoflavone groups were compared with the group receiving milk protein isolate.

28. Hampl *et al.* (2008) reported that short-term consumption of soya had a modest and transitory effect on thyroid parameters, stating that 'some thyroid hormone parameters do correlate with actual isoflavone levels.' Male (n=32) and female (n=54) university students with normal thyroid function consumed unprocessed boiled natural soya beans (2 g/kg bw/day; containing approximately 1.2 – 4.2 mg isoflavones/g dry weight) for 7 days. Levels of TSH, free thyroid hormones, antibodies to TPO and TG, genistein and daidzein were measured in serum collected at baseline, at the end of the treatment period, and one week after completing the soya consumption. Levels of both isoflavones rose at the end of soya consumption and returned to background levels one week later. No significant changes in TSH or free T_3 and T_4 were found in female participants, while in males a significant ($p<0.0001$) transitory increase in TSH was recorded.

29. The effect of 3 years' administration of genistein (54 mg/day) on thyroid function was investigated in osteopenic, postmenopausal women (n=40), who were compared with a control group (n=37) receiving placebo. Thyroid hormones (TSH, free T_3, free T_4) and thyroid-specific autoantibodies (TPO, TG and thyroid microsomal antigen [TMA]) were assessed following the 3-year treatment and were found to be in the normal range and unaffected by time or treatment in either arm (Bitto *et al*., 2010).

30. A study carried out by Li *et al*. (2011) found no association between soya consumption during early pregnancy and development of thyroid dysfunction or autoimmunity. Based on their frequency of soya intake, participants were divided into three groups: frequent (three or more times a week; 18.6%; n=94); "conventional" (> twice per month and < three times per week; 62.6%; n=316); and occasional (≤2 times per month; 18.8%; n=95) consumers. Urinary daidzein and genistein levels were determined in randomly selected frequent, "conventional" and occasional consumers (about 20% of the three groups: n=20, 59 and 16 respectively), and levels of both were significantly higher in frequent consumers. There was no significant difference between the three groups in free T_4, TSH, or TPO antibody levels. No marked difference was found in the prevalence of overt or subclinical hyperthyroidism or overt hypothyroidism. However, there was a non-significant increase in the proportion of subjects with subclinical hypothyroidism among frequent consumers.

31. Effects of isoflavones on thyroid function (blood levels of free T_3 and T_4, TSH, TBG and anti-TPO antibodies) were investigated in a randomised double blind controlled trial among oophorectomised women aged <55 years in India (Mittal *et al*., 2011). Half of the women (n=17) were given a tablet containing 75 mg of isoflavones to be taken orally at bedtime with 150 ml of water, once a day for 12 weeks. The control group (n=17) received a placebo. Participants were advised to avoid food products containing phytoestrogens during the study period. A modest reduction was observed in serum free T_3 levels in the isoflavone group ($p=0.02$), in the absence of any significant effect on other thyroid parameters.

32. In a double-blind, randomised crossover study, healthy postmenopausal women (n=25; mean age 58 years) consumed seaweed capsules (475 µg iodine/day) or placebo (maltodextrose) for 7 weeks, with a three-week washout

period between the treatments (Teas *et al*., 2007). Both treatments were supplemented by powdered soya protein isolate (141.3 mg isoflavones/day) during the 7th week of administration. No changes in serum thyroid hormone concentrations were associated with the isoflavone consumption.

33. Effects of soya consumption on thyroid function have been investigated as a secondary outcome in several other studies. Alekel *et al*. (2014) looked at effects of isoflavone treatment (80 and 120 mg/day) on endometrial thickness, circulating hormones and adverse events in postmenopausal women over a 3 year period. There were no effects on circulating TSH concentrations in either of the treatment groups when compared to baseline levels. Levis *et al.* (2011) reported an absence of significant effects on levels of TSH and antibodies to TPO in menopausal women receiving tablets containing 200 mg isoflavone/day for 2 years. Steinberg *et al.* (2011) reported slightly lower (p=0.052) free T_4 concentrations in menopausal women on isoflavone treatment for 2 years (80 mg/day, n=119; 120 mg/day, n=117) when compared to baseline. No significant changes in TSH concentrations were observed. Nor was there any significant effect of soya foods (approximately 36 mg isoflavones/day) on thyroid hormone status in a study of premenopausal women reported by Zhou *et al*. (2011).

Discussion

34. Since the 2003 COT report, several human studies have provided new data concerning the impact of soya consumption on thyroid function. Some of these investigations suggest that there could be clinically relevant effects in people with treated or sub-clinical hypothyroidism, but the evidence is not entirely consistent.

35. The absence of clear effects on thyroid function in many of the studies may in part reflect differences in the populations studied (e.g. in baseline thyroid status) and in the levels and types of exposure compared (e.g. from diet or supplementation).

36. The FSA-funded research had the most consistent outcomes. In particular, the project designed specifically to assess effects in people with compensated hypothyroidism gave positive results in two separate sub-studies, although only one was statistically significant. Furthermore, in samples of post-menopausal women and men with type 2 diabetes, administration of isoflavone-free soya protein with added isoflavone caused a small but statistically significant reduction in levels of thyroid hormones when compared to treatment with the protein alone.

37. In addition, the finding by Conrad *et al*. (2004) that hypothyroid infants consuming soya formula took longer to respond to thyroxine replacement adds to the suspicion of a problem.

38. If effects of the sort suggested do occur, the most important practical implications are that patients who are known to have borderline hypothyroidism may be at greater risk of requiring thyroxine replacement if they increase their soya intake, and that patients already on such treatment may need a change in dose if

they substantially alter their consumption of soya. These should not present major clinical problems. In particular, patients on thyroid replacement therapy are normally monitored on a regular basis with adjustments to dosage as required. However, endocrinologists should be made aware of the possibility that consumption of soya (including in dietary supplements) may affect thyroid function and response to treatment with thyroxine.

39. In view of the persisting uncertainties, there should be continued monitoring of the scientific literature on this topic. However, since any clinical implications are unlikely to be of major importance, further research in this area need not be a priority for future funding by the Food Standards Agency

Conclusions

40. The Committee considered three FSA-funded studies that provided information about possible effects of soya on thyroid function, and reviewed other relevant research that had emerged since its last published report on the topic in 2003.

41. There are no indications that high intakes of soya impact materially on thyroid function in people who do not already have thyroid disease.

42. However, the evidence that is now available, although not entirely consistent, suggests that higher intake of soya phytoestrogens, either in food or in dietary supplements, may sometimes precipitate a transition to frank disease in people with borderline, compensated hypothyroidism, and may also affect the dose of thyroxine that is needed in patients who are on treatment for hypothyroidism.

43. This should not cause major clinical problems. However, endocrinologists should be made aware of the possibility that consumption of soya (including in dietary supplements) may affect thyroid function and response to treatment with thyroxine.

44. In view of the persisting uncertainties, there should be continued monitoring of the scientific literature on this topic. However, since any clinical implications are unlikely to be of major importance, further research in this area need not be a priority for future funding by the Food Standards Agency.

Secretariat
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Abbreviations

AAP	American Academy of Pediatrics
BfR	German Federal Institute for Risk Assessment
CNS	central nervous system
COT	Committee on Toxicity
CSF	cerebral spinal fluid
EFSA	European Food Safety Authority
ER	oestrogen receptors
FSA	Food Standards Agency
Kd	dissociation constant
NTP	National Toxicology Programe
SD	standard deviation
SPI	soya protein isolate
T_3	triiodothyronine
T_4	thyroxine
TBG	thyroxine binding globulin
TG	thyroglobulin
TMA	thyroid microsomal antigen
TPO	thyroperoxidase
TSH	thyroid stimulating hormone
TTR	transthyretin

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Table 1. Effects on thyroid function reported in human studies following oral exposure to isoflavones

Participants	Dose and duration of soya consumption	Dose (µg/kg bw/day) and duration of levothyroxine treatment	Baseline serum thyroid parameters TSH [mIU/L][1] total T_4/T_3 [nmol/L] free T_4/T_3 [pmol/L] Anti-Tg [U/ml] Anti-TPO [U/ml]	Serum thyroid parameters during/after treatment TSH [mU/L][1] total T_4/T_3 [nmol/L] free T_4/T_3 [pmol/L] Anti-Tg [U/ml] Anti-TPO [U/ml]	Observations	Reference
Children with thyroid dysfunction receiving thyroxine treatment						
Infants diagnosed with congenital hypothyroidism Group A: consuming soya infant formula (n=8; 4M and 4F) Group B: non-soya diet group (n=70; 29M and 41F)	*Soya infant formula* consumed throughout the first year of life; level of isoflavones not specified.	Group A: commenced at a median age of 15 days till 1 year. Starting median dose: 7.4 One year decrease in dose of: 3.3 Group B: commenced at a median age of 17 days till 1 year. Starting median dose: 9.3 One year decrease in dose of: 3.0	Group A: TSH median 428; tT_4 median 29.6 Group B: TSH median 229; tT_4 median 47.6 *Normal ranges: TSH 0.5-5 tT_4 81.1-321.8	After ~ 50 days of treatment Group A:TSH median 42.6 tT_4 median 153 Group B:TSH median 6.6 tT_4 median 188 Time to TSH normalisation Group A: median of 150 days Group B: median of 40 days	4 times longer time to TSH normalisation in soya formula group (p=0.02) 6 times higher TSH measured in soya formula group (p<0.01) 4 times higher % of infants with increased TSH at 4 months of age (p=0.01) and throughout the first year of life in soya formula group CIs not specified	Conrad *et al.*, 2004

Infant girls diagnosed with congenital hypothyroidism <u>Patient 1:</u> consuming soya infant formula <u>Patient 2</u>: consuming soya milk	*Soya infant formula* consumed every 2 hours, 1 hour before thyroxine treatment discontinued at 3 weeks of age *Soya milk* consumed 1 hour before thyroxine treatment between 3 and 4 year of age level of isoflavones in both products not specified	<u>Patient 1:</u> 15 (6 days of age – 3 weeks); 11 (3 – 6 weeks); 8 (6-10 weeks) <u>Patient 2</u>: 6 (3-5 years); 5 (5-5.5 years); 4 (5.5-6 years)	<u>Patient 1</u>: TSH 167 T_4 not specified <u>Patient 2:</u> TSH ~6 tT_4 ~161 *Normal ranges: TSH 1-20 tT_4 141-277 free T_4 9.1-23.8 ~ approximate values read from figure	<u>Patient 1</u>: TSH 216 and tT_4 51 (after 3 weeks); TSH ~25 and tT_4 ~219 (after 6 weeks); TSH ~8 and tT_4 ~203 (after 10 weeks) <u>Patient 2</u>: TSH 248 and free T_4 <5.2 (at 4 years); TSH 1.48 and tT_4 232 (at 5 years); TSH ~1-2 and tT_4 ~148 (at 5.5-6 years) ~ approximate values read from figure	discontinuation of soya-based formula and soya milk and decrease in the dose of thyroxine treatment led to normalisation of thyroid function CIs and p values not specified	Fruzza *et al*., 2012
Adults with normal thyroid function						

Postmenopausal women (45-65 years old) <u>Group A:</u> consuming 80 mg of isoflavones/day <u>Group B</u>: consuming 120 mg of isoflavones/day <u>Group C:</u> consuming placebo	*Isoflavones tablets* provided: Group A: ~0.84-1.83 mg isoflavones/kg bw/day Group B: ~1.35-2.59 mg isoflavones/kg bw/day consumed as 3 compressed tablets/day ~ approximate values calculated based on specified body weights	No treatment.	Not specified for respective groups	Not specified for respective groups	serum TSH measurements as secondary outcome no effects on circulating TSH concentrations in treatment groups when compared to baseline levels	Alekel *et al*., 2014
Osteopenic postmenopausal women <u>Group A</u>: consuming genistein tablets (n=40) <u>Group B</u>: consuming placebo	*Genistein tablets* 54 mg/day duration of treatment was 3 years	No treatment.	<u>Group A</u>: TSH [µg/ml] mean 2.04; fT_4 mean 16.98 fT_3 mean 3.51 Anti-Tg 29.30 Anti-TPO 19.76 <u>Group B:</u>	<u>Group A</u>: TSH [µg/ml] mean 2.02; fT_4 mean 17.50 fT_3 mean 3.41 Anti-Tg 29.00 Anti-TPO 19.35 <u>Group B:</u>	no statistically significant differences in thyroid parameters (all within normal range) between both groups CIs not specified	Bitto *et al*., 2010

(n=37)			TSH [µg/ml] mean 1.69; fT_4 mean 17.37 fT_3 mean 3.60 Anti-Tg 32.90 Anti-TPO 20.33 *Normal ranges: TSH [µg/ml] 0.27-4.2 free T_4 11.96-21.87 free T_3 2.31-5.92	TSH [µg/ml] mean 1.7; fT_4 mean 17.88 fT_3 mean 3.47 Anti-Tg 30.50 Anti-TPO 18.34		
Postmenopausal women not on hormone replacement therapy (64-83 years) <u>Group A:</u> consuming isoflavone supplement (n=22) <u>Group B:</u> consuming placebo (n=16)	*Isoflavone supplement* -30 mg isoflavones consumed three times a day for 6 months participants were asked to maintain normal diet and limit intake of soya-based foods to one serving/week	No treatment.	<u>Group A</u>: TSH mean 3 tT_4 mean 149 tT_3 mean 1.53 <u>Group B:</u> TSH mean 3.35 tT_4 mean 145 tT_3 mean 1.55 *Normal ranges not specified	After 90 days: <u>Group A</u>: TSH mean 3.4 tT_4 mean 149.5 tT_3 mean 1.56 <u>Group B:</u> TSH mean 3.91 tT_4 mean 148 tT_3 mean 1.65 After 180 days: <u>Group A</u>: TSH mean 3.5 tT_4 mean 154.5 tT_3 mean 1.78 <u>Group B:</u>	no statistically significant differences in thyroid parameters (all within normal range) between both groups CIs not specified	Bruce *et al.,* 2003

				TSH mean 3.63 tT_4 mean 154 tT_3 mean 1.75		
Healthy men (n=35; 20-40 years) <u>Arm 1:</u> Milk Protein Isolate (MPI) <u>Arm 2</u>: Low-isoflavone Soya Protein Isolate (low-iso SPI) <u>Arm 3:</u> High-isoflavone Soya Protein Isolate (high-iso SPI)	*Low-iso SPI* mean ± SD 1.64 ± 0.19 mg isoflavones/day *High-iso SPI* mean ± SD 61.7 ± 7.4 mg isoflavones/day all participants took part in 3 arms (57 days each) separated by 28 days of washout period participants were asked to maintain normal diet and limit intake of soya-based foods	No treatment	<u>Arm 1:</u> TSH mean 2.14 (95% CI 1.97-2.33) tT_4 mean 80.4 fT_4 mean 17.3 tT_3 mean 1.78 fT_3 mean 4.76 <u>Arm 2</u>: TSH mean 2.14 (95% CI 1.97-2.33) tT_4 mean 79.2 fT_4 mean 17.1 tT_3 mean 1.76 fT_3 mean 4.79 <u>Arm 3</u>: TSH mean 2.11 (95% CI 1.94-2.30) tT_4 mean 78.8 fT_4 mean 16.9 tT_3 mean 1.76 fT_3 mean 4.77	After 29 days: <u>Arm 1:</u> TSH mean 2.04 (95% CI 1.87-2.22) tT_4 mean 78.9 fT_4 mean 16.3 tT_3 mean 1.79 fT_3 mean 4.9 <u>Arm 2:</u> TSH mean 1.82 (95% CI 1.67-1.98) tT_4 mean 75.5 fT_4 mean 16.3 tT_3 mean 1.75 fT_3 mean 4.69 <u>Arm 3</u>: TSH mean 1.91 (95% CI 1.76-2.08) tT_4 mean 78.5 fT_4 mean 16.6 tT_3 mean 1.80 fT_3 mean 4.75 After 57 days: <u>Arm 1</u>: TSH mean 1.99 (95% CI 1.82-2.18)	no significant differences in serum concentrations of thyroid parameters among groups during the study urinary excretion of isoflavones was 19 (daidzein) and 18 (genistein) folds higher in Arm 3 when compared to Arm 2, and 26 and 28 folds higher when compared to Arm 1 respectively ($p<0.0001$ for all comparisons).	Dillingham *et al.*, 2007

			*Normal ranges not specified	tT_4 mean 79.7 fT_4 mean 16.9 tT_3 mean 1.77 fT_3 mean 4.7 <u>Arm 2:</u> TSH mean 1.90 (95% CI 1.73-2.08) tT_4 mean 80 fT_4 mean 16.8 tT_3 mean 1.83 fT_3 mean 4.92 <u>Arm 3:</u> TSH mean 2.02 (95% CI 1.84-2.21) tT_4 mean 80.5 fT_4 mean 17.1 tT_3 mean 1.77 fT_3 mean 4.80		
University students without overt thyroid disease (n=86; 32M and 54F; 18-25 years old)	*Natural soya beans* unprocessed, boiled 2 g of soya beans/kg bw/day consumed for 7 days	No treatment.	<u>Men:</u> TSH ~1.4 fT_4 ~12.25 fT_3 ~5.25 <u>Women:</u> TSH ~1.5 fT_4 ~11.5 fT_3 ~4.8 *Normal ranges:	<u>Men:</u> TSH ~1.8; fT_4 ~12.35; fT_3 ~5 (after 7 days of treatment); TSH ~1.6; fT_4 ~11.9; fT_3 ~5.12 (7 days after treatment termination)	levels of isoflavones significantly increased following soya consumption: for genistein (by 4 folds in men and ~3 folds in women) and for daidzein (by 8 folds in men and by 4 folds in women) ($p<0.0001$). Return to nearly initial values 7 days	Hampl *et al.*, 2008

	isoflavone content: approximately 1.2 to 4.2 mg per 1 g of dry weight		TSH 0.3-4.2 ~ approximate values read from figure	<u>Women:</u> TSH ~1.55; fT_4 ~11.7; fT_3 ~4.65 (after 7 days of treatment); TSH ~1.46; fT_4 ~11.8; fT_3 ~4.8 (7 days after treatment termination)	after treatment termination. no significant changes in thyroid parameters were found in women a significant increase in TSH ($p<0.0001$) in men following soya consumption CIs not specified	
Women within 5 years of menopause (45-60 years old) <u>Group A</u>: consuming soya isoflavone tablets (n=122) <u>Group B:</u> consuming placebo (n=126)	*Soya isoflavone tablets* 200 mg of isoflavones (91 mg genistein and 103 mg daidzein) corresponding to 2.05-4.50 mg/kg bw/day taken as 4 tablets daily (50 mg each) for 2 years	No treatment.	Not specified.	<u>Group A:</u> TSH increased by 0.32 (after 1 year treatment) and by 0.04 (after 2 year treatrment) <u>Group B</u>: TSH decreased by 0.15 (after 1 year treatment) and by 0.61 (after 2 year treatment)	thyroid parameters as secondary outcome no effects on TSH concentrations in treatment groups when compared to baseline levels mean total urinary isoflavones increased significantly by 56.5 pmol/µl in Group A ($p<0.001$), and by 2.9 pmol/µl in Group B after 2 years of treatment.	Levis *et al.,* 2011
Women in early stages of pregnancy	*Soya consumption*:	No treatment.	Not measured	<u>Group A</u>: TSH mean 2.38	urinary levels of genistein and daidzein (measured	Li *et al*., 2011

(n=505) Group A: frequent soya consumers (n=94) Group B: conventional soya consumers (n=316) Group C: occasional soya consumers (n=95)	frequent – 3 or more times/week conventional – 2 times/month to 3 times/week occasional – 2 or fewer times/month type of food and level of isoflavones not specified		*Normal ranges: TSH 0.13-3.93 fT_4 12-23.34 fT_3 3.46-7.70	fT_4 mean 17.93 Group B: TSH mean 2.12 fT_4 mean 18.15 Group C: TSH mean 2.35 fT_4 mean 18.20	in 20% of participants) were significantly lower in conventional (n=59) and occasional (n=16) consumers when compared with frequent (n=20) consumers ($p<0.01$) no significant difference in TSH and free T_4 between all groups CIs not specified	
Oophorectomised women (<55 years old) Group A: consuming isoflavone tablet (n=17) Group B: consuming placebo tablet (n=17)	*Isoflavone tablet* contained 75 mg of soya isoflavones consumed once a day for 12 weeks participants were asked to avoid soya-based foods	No treatment.	Group A: TSH mean 1.87 fT_4 mean 13.88 fT_3 mean 4.05 Anti-TPO mean 42.06 Group B: TSH mean 2.04 fT_4 mean 13.45 fT_3 mean 4.21 Anti-TPO mean 98.25 *Normal ranges: TSH 0.5-5.5	Group A: TSH mean 1.5; fT_4 mean 13.88; fT_3 mean 4.12; Anti-TPO mean 61.42 (after 6 weeks); TSH mean 3.28; fT_4 mean 13.61; fT_3 mean 3.76; Anti-TPO mean 63.75 (after 12 weeks) Group B: TSH mean 1.83; fT_4 mean 13.35; fT_3	significant decrease in free T_3 levels following treatment with isoflavones in group A ($p=0.02$) no other significant changes in thyroid parameters after 12 weeks of treatment between the groups CIs not specified	Mittal *et al.*, 2011

				mean 4.12; Anti-TPO mean 134.58 (after 6 weeks); TSH mean 2.72; fT_4 mean 13.55; fT_3 mean 4.07; Anti-TPO mean 131.53 (after 12 weeks);		
Menopausal women (40-60 years old) <u>Group A:</u> consuming 80 mg of isoflavones/day <u>Group B</u>: consuming 120 mg of isoflavones/day <u>Group C:</u> consuming placebo	*Isoflavones tablets* provided: <u>Group A</u>: mean ~1.16 mg isoflavones/kg bw/day <u>Group B</u>: mean ~1.77 mg isoflavones/kg bw/day ~ approximate values calculated based on specified body weights intake of soya food limited to one serving/week	No treatment.	<u>Group A:</u> TSH 2.2; fT_4 15.44 (n=135) <u>Group B</u>: TSH 2.4; fT_4 15.44 (n=134) <u>Group C</u>: TSH 2.8; fT_4 14.16 (n=134) *Normal ranges: TSH 0.35-5.5 fT_4 10.29-23.16	<u>Group A</u>: TSH 2.5; fT_4 15.44 (after 1 year treatment, n=122) and TSH 2.4; fT_4 14.16 (after 2 years of treatment, n=119) <u>Group B</u>: TSH 2.6; fT_4 15.44 (after 1 year of treatment, n=123) and TSH 2.2; fT_4 14.16 (after 2 years of treatment, n=117) <u>Group C:</u> TSH 2.6; fT_4 14.16 (after 1 year of treatment, n=128) and TSH 2.4; fT_4	thyroid parameters as secondary outcome slightly lower (p=0.052) free T_4 levels in Group A and B when compared to placebo no significant changes in TSH concentrations after 1 and 2 years of treatment between the groups CIs not specified	Steinberg *et al*., 2011

				15.44 (after 2 years of treatment, n=126)		
Postmenopausal women (mean age 58 years) <u>Arm 1:</u> consuming seaweed capsules for 7 weeks (+high isoflavone powder in the 7th week of treatment) <u>Arm 2:</u> consuming placebo capsules for 7 weeks (+high isoflavone powder in the 7th week of treatment)	*High isoflavones powder* 2 mg of isoflavones/kg bw/day consumed once a day for 7 days in the last week of treatment all participants took part in 2 arms (7 weeks each) separated by 3 week washout period	No treatment	Not specified	<u>Arm 1</u>: TSH mean 2.19; tT_4 mean 85; tT_3 mean 1.97 (after 6 weeks with seaweed only); TSH mean 1.94; tT_4 mean 86; tT_3 mean 1.94 (after 7 weeks when isoflavones were added for one week) <u>Arm 2</u>: TSH mean 1.69; tT_4 mean 85; tT_3 mean 1.91 (after 6 weeks with placebo only); TSH mean 1.64; tT_4 mean 87.5; tT_3 mean 1.93 (after 7 weeks when isoflavones were added for one week)	seaweed consumption significantly increased levels of TSH and urinary iodine excretion ($p<0.01$). This effect was not altered by isoflavone ingestion no significant changes in thyroid parameters following isoflavone consumption CIs not specified	Teas *et al*., 2007
Premenopausal women (18-28 years old)	*Soya foods* soya protein	No treatment.	<u>Group A:</u> TSH ~0.027 fT_4 15.83	<u>Group A:</u> TSH ~0.029 fT_4 15.70	thyroid parameters as secondary outcome	Zhou *et al*., 2011

Group A: consuming soya foods (n=31) Group B: consuming animal foods (n=32)	content ranged from 18 to 22 g/day isoflavone content was approximately 36 mg/day 2 to 3 servings per day during 10 weeks period participants were asked to restrict any additional soya foods and limit animal foods to one serving/day		Group B: TSH ~0.030 fT_4 15.70 ~ approximate values read from figure *Normal ranges not specified	Group B: TSH ~0.026 fT_4 15.44 ~ approximate values read from figure	no significant changes in thyroid parameters following soya food consumption significant difference in level of TSH between two groups ($p<0.05$) mean total urinary isoflavones increased significantly by 16 folds in Group A ($p=0.016$), and decreased by 1.4 fold in Group B CIs not specified	
Food Standards Agency-funded research "The effect of soya phytoestrogen supplementation on thyroid status and cardiovascular risk markers in patients with subclinical hypothyroidism"						
PART ONE						University of Hull 2005-2015 PART ONE published by Sathyapalan *et al.*, 2011
Patients with subclinical hypothyroidism (n=60; 8M and 52F; 44-70 years old) Arm 1: consumed 2 mg	*Phytoestrogen material* 54% genistein, 35% daidzein and 12% glycitein participants	Thyroxine treatment was commenced after diagnosis with overt hypothyroidism	Arm 1: TSH mean 7.8 fT_4 mean 11.8 fT_3 mean 4.0 Arm 2: TSH mean 7.9 fT_4 mean 12.2	Arm 1: TSH mean 7.5; fT_4 mean 11.9; fT_3 mean 4.4 (after 3 months) Arm 2: TSH mean 8.4; fT_4	6 patients (10%; all females) developed overt hypothyroidism (defined as TSH>4.7 mU/L and fT_4 <9 pmol/L) after treatment with 16 mg isoflavones	

phytoestrogen with 30 g soya protein powder <u>Arm 2:</u> consumed 16 mg phytoestrogen with 30 g soya protein powder	were asked to avoid soya-based foods all participants took part in 2 arms (8 weeks each) separated by 8 week washout period		fT_3 mean 4.2 *Normal ranges: TSH 0.5-4.7 fT_4 9-24 fT_3 2.5-5.3	mean 11.3; fT_3 mean 4.3 (after 3 months)	Anti-TPO were positive (>75 U/ml) in 38 (63.3%) patients no significant changes in thyroid parameters between two arms levels of isoflavones significantly increased following phytoestrogen supplementation: for genistein (by 4 folds in arm 1 and 19 folds in arm 2) and for daidzein (by 5.5 folds in arm 1 and 12 folds in arm 2) ($p<0.01$)	
PART TWO						
Patients with subclinical hypothyroidism (n=34; 17M and 17F; 23-80 years old) <u>Arm 1:</u> consumed 30 g soya protein powder (phytoestrogen free)	*Phytoestrogen material* 54% genistein, 35% daidzein and 12% glycitein participants were asked to avoid soya-based foods	Thyroxine treatment was commenced after diagnosis with overt hypothyroidism	<u>Arm 1</u>: TSH mean 5.0 fT_4 mean 12.2 fT_3 mean 4.6 <u>Arm 2</u>: TSH mean 5.3 fT_4 mean 11.3 fT_3 mean 4.7	<u>Arm 1</u>: TSH mean 5.2; fT_4 mean 12.0; fT_3 mean 4.3 (after 2 months) <u>Arm 2</u>: TSH mean 5.5; fT_4 mean 11.9; fT_3 mean 4.7 (after 2 months)	2 patients (6%; 1M, 1F) developed overt hypothyroidism (defined as TSH>4.7 mU/L and fT_4 <9 pmol/L) after treatment with 66 mg phytoestrogens Anti-TPO were positive (>75 U/ml) in 17 (50%) patients	

Arm 2: consumed 66 mg phytoestrogen with 30 g soya protein powder	all participants took part in 2 arms (8 weeks each) separated by 8 week washout period		*Normal ranges: TSH 0.5-4.7 fT_4 9-24 fT_3 2.5-5.3		statistically significant increase in TSH (p=0.004) and decrease in fT_3 (p=0.03) in males after treatment with 66 mg phytoestrogens no significant changes in thyroid parameters between two arms levels of isoflavones significantly increased following 66 mg phytoestrogen supplementation: by 19 folds for genistein and by 8 folds for daidzein (p=0.00) CIs not specified	
PART THREE						
Patients with subclinical hypothyroidism (n=42; 16M and 26F; 23-80 years old) Arm 1: consumed 30 g soya	all participants took part in 2 arms (8 weeks each) separated by 8 week washout period	No treatment.	Arm 1: TSH 5.70; fT_4 12.60; fT_3 4.44 Arm 2: TSH 5.93; fT_4 12.11;	Arm 1: TSH 5.86 (95% CI: -0.06-0.16); fT_4 11.06 (95% CI: -0.96-0.5); fT_3 4.37 (95% CI: -0.12-0.11)(after 3 months)	none of the patients developed overt hypothyroidism during the study period no significant changes in thyroid parameters were observed after the	

protein powder (phytoestrogen free) <u>Arm 2:</u> consumed 30 g casein protein powder			fT_3 4.40 *Normal ranges: TSH 0.5-4.7 fT_4 9-24 fT_3 2.5-5.3	<u>Arm 2:</u> TSH 5.71 (95% CI: -0.71-0.60); fT_4 11.93 (95% CI: -0.36-0.12); fT_3 4.43 (95% CI: -0.14-0.20) (after 3 months)	treatment	
Food Standards Agency-funded research "A double blind placebo controlled parallel trial of soya isoflavones on markers of bone turnover in females in early menopause"						
Women within two years of the onset on menopause (50-55 years old) <u>Group A:</u> consumed 30 g soya protein powder with 66 mg phytoestrogen (n=101) <u>Group B:</u> consumed 30 g soya protein powder (phytoestrogen free) (n=99)	*Soya protein powder and isoflavones* consumed twice a day as a 15 g bar containing 33 mg isoflavones for 6 months		<u>Group A</u>: TSH median 1.5 fT_4 median 13 fT_3 median 4.6 <u>Group B:</u> TSH median 1.6 fT_4 median 13 fT_3 median 4.7	<u>Group A:</u> TSH ~2.9; fT_4 ~12 (after 3 months); TSH ~2.9; fT_4 ~11 (after 6 months) <u>Group B</u>: TSH ~1.65; fT_4 ~13 (after 3 months); TSH ~1.6; fT_4 ~13 (after 6 months) ~ approximate median values read from figure	significant increase (although within normal ranges) in AUC for TSH (2.34 vs 1.71 mU/L; p=0.004; 95% CI: 0.18-0.95) and significant decrease in AUC for fT_4 (12.2 vs 13.1 pmol/L; p=0.02; 95% CI: -1.8- -0.1) with Group A compared to Group B significant increase in AUC for daidzein (22.6 vs 4.3 ng/ml; p<0.001; 95% CI:11.3-25.3) and genistein (89.1 vs 8.4 ng/ml; p<0.001; 95% CI: 56.3-104.8) following consumption of	University of Hull 2010-2014

					phytoestrogens when Group A compared to Group B	
Food Standards Agency-funded research "A double blind placebo controlled parallel trial of soya phytoestrogens in patients with compensated hypogonadism"						
Men with type 2 diabetes and subclinical hypogonadism (45-75 years old) <u>Group A:</u> consumed 30 g soya protein powder with 66 mg isoflavones (n=107) <u>Group B:</u> consumed 30 g soya protein powder (isoflavone free) (n=103)	*Soya protein powder and isoflavones* consumed twice a day as a 15 g bar containing 33 mg isoflavones for 3 months participants were asked to avoid soya-based foods	No treatment.	<u>Group A</u>: TSH median 1.82 fT_4 median 12.62 fT_3 median 4.66 <u>Group B:</u> TSH median 1.81 fT_4 median 13.08 fT_3 median 4.64	<u>Group A</u>: TSH median 3.28; fT_4 median 11.08; fT_3 median 4.71 (after 3 months) <u>Group B:</u> TSH median 1.97; fT_4 median 12.71; fT_3 median 4.55 (after 3 months)	significant increase (although within normal ranges) in TSH level ($p<0.01$; 95% CI: -1.63- -1.28) and decrease in free T_4 ($p<0.01$; 95% CI: 0.96-2.12) following treatment with 66 mg isoflavones levels of isoflavones significantly increased following 66 mg isoflavone supplementation: by 20 folds for genistein and by 13 folds for daidzein ($p<0.01$)	University of Hull 2009-2015

[1] Except Bitto *et al*., 2010 where different technique was used to measure TSH, which levels were reported as µg/ml as opposed to mIU/L

TSH – thyroid stimulating hormone; fT_3 – free triiodothyronine; tT_3 – total T_3; fT_4 – free thyroxine; tT_4 – total T_4; CI – confidence interval; Anti-Tg – thyroglobulin autoantibodies; Anti-TPO – thyroperoxidase autoantibodies; SPI – soya protein isolate; MPI – milk protein isolate; AUC – area under the curve

Appendix 1

Search strategy

Websites of international authorities/advisory bodies interrogated

- COT
- EFSA
- FSA
- IARC
- JECFA

Scientific literature search

PubMed was interrogated to identify publications published until November 2014 and relevant to effects of soya consumption on thyroid status. Specific search terms and the number of hits associated with those queries are listed below. The list below does not take into account overlap of search results between search terms. These hits have been further refined via interrogation of the abstracts and the articles themselves, excluding articles that were investigating effects of soya-based food products/isoflavones/phytoestrogens in combination with other compounds, in individuals with health problems not relevant to thyroid, in individuals with concomitant illnesses, studies in animals and in vitro studies, studies mentioned in the 2003 COT report. A reasonable number of relevant studies investigating thyroid effects in humans have been found. Therefore they have been the only ones included in this statement. The final list of articles used in this document can be found in the references section.

Specific search terms	Hits in Pubmed
Flavonoids AND hypothyroidism	27
Flavonoids AND subclinical hypothyroidism	2
Genistein/Daidzein/Glycitein AND hypothyroidism	4/2/0
Genistein/Daidzein/Glycitein AND subclinical hypothyroidism	1/0/0
Genistein/Daidzein/Glycitein AND thyroid	93/30/1
Isoflavones AND hypothyroidism	20
Isoflavones AND subclinical hypothyroidism	2
Isoflavones AND thyroid	143
Phytoestrogens AND hypothyroidism	6

Phytoestrogens AND subclinical hypothyroidism	2
Phytoestrogens AND thyroid	98
Soy/Soya AND hypothyroidism	22/22
Soy/Soya AND subclinical hypothyroidism	1/1
Soy/Soya AND thyroid	124/134
Soy/Soya protein AND hypothyroidism	7/5
Soy/Soya protein AND subclinical hypothyroidism	1/0
Soy/Soya protein AND thyroid	71/52